AD-A176 898 THE USE OF HYPERTONIC SOLUTIONS TO RESUSCITATE ANIMALS FROM HYPOVOLEMIC SHOCK(U) CALIFORNIA UNIV DAVIS J W HOLCROFT 18 MAR 86 DAMD17-83-C-3175 1/1

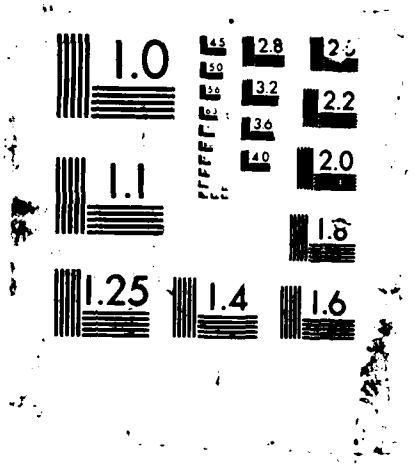
UNCLASSIFIED F/G 6/15 NL

END
DATE FILMED
8 7

[redacted]

Appendix
#1

Circulatory Shock 13:149-159 (1984)


1.0
1.1
1.25
1.4
1.6
2.8
3.2
3.6
4.0
2.2
2.0
1.8

AD-A176 898

[illegible]

ANNUAL/FINAL REPORT

[illegible]

JAMES W. HOLCROFT, M.D.

Supported by

U.S. ARMY MEDICAL RESEARCH AND DEVELOPMENT COMMAND
Fort Detrick, Frederick, Maryland 21701-5012

[illegible]

DTIC
ELECTE
S D
E

SECURITY CLASSIFICATION OF THIS PAGE

REPORT DOCUMENTATION PAGE

1a. REPORT SECURITY CLASSIFICATION: UNCLASSIFIED	1b. RESTRICTIVE MARKINGS
2a. SECURITY CLASSIFICATION AUTHORITY	3. DISTRIBUTION/AVAILABILITY OF REPORT: Approved for public release; distribution unlimited
2b. DECLASSIFICATION/DOWNGRADING SCHEDULE	
4. PERFORMING ORGANIZATION REPORT NUMBER(S)	5. MONITORING ORGANIZATION REPORT NUMBER(S)
6a. NAME OF PERFORMING ORGANIZATION: University of California, Davis; 6b. OFFICE SYMBOL (If applicable)	7a. NAME OF MONITORING ORGANIZATION
6c. ADDRESS (City, State, and ZIP Code): Sacramento, California 95616	7b. ADDRESS (City, State, and ZIP Code)
8a. NAME OF FUNDING/SPONSORING ORGANIZATION: U.S. Army Medical Research & Development Command; 8b. OFFICE SYMBOL (If applicable)	9. PROCUREMENT INSTRUMENT IDENTIFICATION NUMBER: DAMD17-83-C-3175
8c. ADDRESS (City, State, and ZIP Code): Fort Detrick, Frederick, Maryland 21701-5012	10. SOURCE OF FUNDING NUMBERS: PROGRAM ELEMENT NO. 62772A; PROJECT NO. 3S1-62772A874; TASK NO. AB; WORK UNIT ACCESSION NO. 261

11. TITLE (Include Security Classification)

(U) THE USE OF HYPERTONIC SOLUTIONS TO RESUSCITATE ANIMALS FROM HYPOVOLEMIC SHOCK

12. PERSONAL AUTHOR(S)
Holcroft, J.W.

13a. TYPE OF REPORT	13b. TIME COVERED	14. DATE OF REPORT (Year, Month, Day)	15. PAGE COUNT
Final Annual/ Final *	FROM 7/01/83 TO 09/30/85	March 18, 1986	14

16. SUPPLEMENTARY NOTATION
*Annual for the period Jul 84-Sep 85
Final for the period Jul 83-Sep 85

17. *CONT'D COSATI CODES		
FIELD	GROUP	SUB-GROUP
06	05	
06	11	

18. SUBJECT TERMS (Continue on reverse if necessary and identify by block number)
Hypovolemic shock; hemorrhagic shock; hemorrhagic hypotension; resuscitation; hyperosmotic solutions; hypertonic sodium chloride; hypertonic saline; hyperoncotic *Cont.

19. ABSTRACT (Continue on reverse if necessary and identify by block number)

The purpose of this contract was to evaluate small volume resuscitation of animals subjected to moderately severe hemorrhagic shock. The stimulus for this study was the observation that many soldiers who die preventable deaths after suffering injuries die from uncontrolled hemorrhagic shock. Resuscitation of soldiers in the field is difficult, in part, because of the necessity to infuse large volumes of isotonic resuscitative solutions. This proposal evaluated the use of very small volumes of hyperosmotic solutions. The proposal began by evaluating different compositions for hyperosmotic solutions and ended with an evaluation of the solution that appeared the most promising. We found that 2,400 milliosmolar sodium chloride with 6% Dextran 70 was highly effective in resuscitating animals that had lost up to 60% of their blood volume. The amount of solution necessary to restore and maintain cardiac index and blood pressure was on the order of 1/5 the amount of blood loss from the hemorrhagic insult. These solutions appear particularly promising because they are inexpensive, require no cross matching, *Cont.

20. DISTRIBUTION/AVAILABILITY OF ABSTRACT: ☒ UNCLASSIFIED/UNLIMITED ☐ SAME AS RPT. ☐ DTIC USERS	21. ABSTRACT SECURITY CLASSIFICATION: UNCLASSIFIED
22a. NAME OF RESPONSIBLE INDIVIDUAL: Mrs. Judy Pawlus	22b. TELEPHONE (Include Area Code): 301/663-7325; 22c. OFFICE SYMBOL: SGRD-RMI-S

DD FORM 1473, 84 MAR — 83 APR edition may be used until exhausted. All other editions are obsolete. — SECURITY CLASSIFICATION OF THIS PAGE

17. CONT'D COSATI CODES

FIELD	GROUP
06	01

18. (Continued) Subject Terms

solutions; battlefield casualties; small volume resuscitation; cardiac output; cardiac index; blood pressure; heart rate; urine output; arterial blood gasses; acid base balance; skeletal muscle membrane potentials; intracellular and extracellular volumes; Dextran; albumin; mannitol; sodium bicarbonate; sheep; rats.

19. (continued) Abstract

are effective when used in small volumes, are easy to store, and should keep indefinitely as they will not support bacterial growth.

AD _____

THE USE OF HYPERTONIC SOLUTIONS TO RESUSCITATE ANIMALS FROM HYPOVOLEMIC SHOCK

ANNUAL/FINAL REPORT

MARCH 18, 1986

JAMES W. HOLCROFT, M.D.

Supported by

U.S. ARMY MEDICAL RESEARCH AND DEVELOPMENT COMMAND
Fort Detrick, Frederick, Maryland 21701-5012

Contract No. DAMD17-83-C-3175

University of California
Davis, California 95616

SUMMARY

Hemorrhagic shock has remained one of the main causes of death in war for the last century. Effective resuscitation requires infusion of large volumes of fluid, enough to restore both the vascular volume and the extracellular space. Effective field resuscitation has been limited because it is difficult to carry large volumes of fluid into battle areas and because the time required to infuse large volumes of fluid is limited under battlefield conditions. The purpose of this study, as initially proposed, was to evaluate the use of extremely hypertonic solutions in resuscitating animals from hypovolemic shock. The goal was to demonstrate, if possible, that such fluids could, indeed, effectively resuscitate hypovolemic animals. Another goal of the proposal was to demonstrate which of the many possible hypertonic solutions is most effective in such resuscitation.

In brief, we have been able to fulfill all of the objectives listed in the initial proposal. We have found that extremely hypertonic sodium chloride solutions are highly effective in the initial resuscitation of animals from hypovolemic shock, and we have found that administering these solutions in combination with Dextran makes the initial beneficial effects last for some 30 minutes or even longer. These hypertonic solutions appear very promising in resuscitating soldiers injured in battle. They may also be of considerable value in resuscitating civilians injured in automobile accidents or by other means.

FOREWORD

For part of these studies, we used Dextran solutions, manufactured by Pharmacia. Citations of this trade name and commercial organization do not constitute an official Department of the Army endorsement or approval of the products or services of this organization.

In conducting the research described in this report, the investigator(s) adhered to the "Guide for the Care and Use of Laboratory Animals," prepared by the Committee on Care and Use of Laboratory Animals of the Institute of Laboratory Animal Resources, National Research Council (DHEW Publication No. (NIH) 78-23, Revised 1978).

TABLE OF CONTENTS

BODY OF REPORT

Background

One of the main causes of death in injured soldiers during the past century has been hemorrhagic shock. Resuscitation of hemorrhagic shock requires infusion of large volumes of conventional fluids. This led to difficulties in battlefield conditions because it is impossible to carry adequate amounts of isotonic solutions into a combat zone and because time is limited and infusion of large volumes of fluid requires too much time if a medic has to care for many injured soldiers. This problem led to the proposal initially suggested for this contract, namely, that infusion of very small volumes of very hypertonic solutions might successfully resuscitate animals (and possibly human beings) from hemorrhagic shock. We had had some experience with using very hypertonic solutions in animal work before our initial proposal was made, and the idea of using very hypertonic solutions in resuscitating animals from shock goes back at least 40 years. (1-4)

The purpose of this contract proposal was to determine if, indeed, very hypertonic solutions are effective in resuscitating animals from hemorrhagic shock and to determine how best to constitute such solutions so as to make them the most effective.

Results and Discussion

The work proposed in the contract has resulted in several publications, including the following: "Small Volume Resuscitation with Hypertonic Saline (2,400 mOsm/Liter) during Hemorrhagic Shock", "Infusion of Very Hypertonic Saline to Bled Rats: Membrane Potentials and Fluid Shifts", "A Comparison of Several Hypertonic Solutions for Resuscitation of Bled Sheep", and "Small Volume Resuscitation with Hypertonic Saline Dextran Solution", which was presented at the recent meeting of the Society of University Surgeons and has been submitted for publication in **Surgery**. Copies of these publications are included with this report as appendices, but to facilitate review I will summarize those studies.

In the first study by Nakayama, et al, published in **Circulatory Shock** in 1984, we documented that small volume resuscitation, using either normal saline or hypertonic saline (2,400 mOsm/Liter) following hemorrhagic shock, resuscitated unanesthetized sheep for a period of some 15-30 minutes. This resuscitation was associated with an immediate and marked improvement in blood pressure, cardiac output, pulse and filling

pressures of the heart. Some of the improvement was caused by an increase in the plasma volume. Some of the improvement, apparently, was not related directly to the increase in plasma volume but, at that time, we did not know the mechanisms for the response. The response was dramatic enough and good enough so that the animals did survive the shock insult and the subsequent resuscitation; at the same time, the response was not as long lasting as we would have liked.

This work lead to the study by Smith, et al, that was published in the **Journal of Surgical Research** in 1985. That study searched for the best solution to use in resuscitating sheep from hemorrhagic shock. Several different solutions were used and several different osmolalities. These solutions included hypertonic sodium chloride, hypertonic sodium bicarbonate, hypertonic sodium chloride/sodium acetate, hypertonic sodium chloride/mannitol, hypertonic glucose, and hypertonic sodium chloride/6% Dextran 70. All of the hypertonic solutions resuscitated the animals well for several minutes; the hypertonic sodium chloride maintained resuscitation for 15-30 minutes; the solution of hypertonic sodium chloride and 6% Dextran 70, however, maintained resuscitation indefinitely. The combination of the sodium chloride and Dextran was better than either of the component parts. The animals' osmolality increased some 10% with resuscitation and the plasma sodium concentration increased some 7%. The animals had no ill effects from these increases and the sheep tolerated the resuscitation extremely well. It appeared, on the basis of these studies, that many hypertonic solutions are effective in giving an initial beneficial response in resuscitating animals from hemorrhagic shock, but that the combination of the hypertonic crystalloid solution with the hyperoncotic colloid solution was best for maintenance of the beneficial effects.

This study then lead to a study in which the hypertonic sodium chloride/6% Dextran 70 solution was used in an experimental situation that was designed to mimic the field situation. Animals were subjected to hemorrhagic shock and then resuscitated with small volumes of the hypertonic sodium chloride/Dextran solution. The resuscitation consisted only of the solution and nothing else for a total of 30 minutes, at which time the animals were then given isotonic solutions in quantities sufficient to maintain their cardiac indices at baseline levels. The 30 minute time, in which only the hypertonic solutions were used, mimiced the ambulance transport time that a patient is likely to undergo after an injury; the resuscitation with isotonic solutions mimiced the resuscitation once the patient reached a hospital facility with unlimited supplies of solutions. The hypertonic solutions fully restored all cardiovascular measurements to baseline levels, including blood pressure, heart rate, right atrial pressure, total peripheral resistance, stroke volume, pulmonary artery pressure,

respiratory rate, blood gases, and plasma lactate concentrations. Very little volume was needed in the hospital setting scenario to complete the resuscitation in the animals given the hypertonic saline/Dextran combination.

Lastly, a study was conducted on the effects of hypertonic saline on resting skeletal muscle transmembrane potentials. Rats were bled to produce a moderate degree of hemorrhagic shock until their resting skeletal muscle transmembrane potentials deteriorated to -65 mV. Hypertonic saline was used for resuscitation. This resuscitation restored the membrane potentials to their normal values of some -82 mV and also restored cellular water and cellular sodium and chloride contents to baseline levels. That is, the hypertonic saline solution reversed some of the cellular abnormalities that were induced by the hemorrhagic shock.

Conclusion

In summary, these studies have culminated in the evaluation of a solution that is effective in resuscitating unanesthetized sheep from moderate degrees of hemorrhagic shock. The solution works well in very small volumes - volumes that are small enough so that they could easily be carried into a battlefield by medics. The volumes used were on the order of 1/5 of that required when isotonic solutions are used. We are in the process of evaluating these solutions in more detail and in attempting to determine some of the mechanisms of actions of these solutions.

Recommendations

These results are promising and could well lead to more effective resuscitation of soldiers that are injured in combat. We believe that the solutions should be evaluated further. The mechanism of their actions should be further delineated. Human studies should be begun.

LITERATURE CITED

1. Nakayama, S., Sibley, L., Gunther, R., Holcroft, J., Kramer, G.: Small-Volume Resuscitation with Hypertonic Saline (2,400 mOsm/Liter) during Hemorrhagic Shock. Circulatory Shock 13:149-159, 1984.

2. Nakayama, S., Kramer, G., Carlsen, R., Holcroft, J.: Infusion of Very Hypertonic Saline to Bled Rats: Membrane Potentials and Fluid Shifts. Journal of Surgical Research 38:180-186, 1985.

3. Smith, G., Kramer, G., Perron, P., Nakayama, S., Gunther, R., Holcroft, J.: A Comparison of Several Hypertonic Solutions for Resuscitation of Bled Sheep. Journal of Surgical Research 39:517-528, 1985.

4. Kramer, G., Perron, P., Lindsey, D., Ho, H., Gunther, R., Boyle, W., Holcroft, J. Submitted to: Surgery, 1986.

APPENDICES

#1 - #4

Appendix
#1

Circulatory Shock 13:149–159 (1984)

Small-Volume Resuscitation With Hypertonic Saline (2,400 mOsm/Liter) During Hemorrhagic Shock

Shin-ichi Nakayama, Lillian Sibley, Robert A. Gunther, James W. Holcroft, and George C. Kramer

Departments of Surgery and Human Physiology, University of California, Davis, and Second Department of Surgery, Kyushu University, School of Medicine, Fukuoka, Japan

We compared small-volume resuscitation using either normal saline or hypertonic saline (2400 mOsm/liter) during hemorrhagic hypotension. Six unanesthetized sheep were bled to 50 mm Hg mean arterial pressure and maintained for 2 h. During this shock period cardiac output decreased 40–50% of baseline, while total peripheral resistance increased 20–30%. Then the response to a bolus injection of either hypertonic saline or normal saline, randomly chosen, was studied for an additional 2 h. The volume injected was 145–175 ml, equal to 10% of total shed blood volume. After data collection all shed blood was returned. Several days later, the protocol was repeated on each sheep with the alternate solution. After hypertonic saline the mean arterial pressure increased 48 mm Hg to 83% of control; with normal saline, mean arterial pressure increased 26 mm Hg. Cardiac output recovered to 95% of control immediately after infusion of hypertonic saline, while no significant increase was observed with normal saline. Ten minutes after injection of hypertonic saline, plasma volume increased ~360 ml, but with normal saline no increase was observed. We conclude that small-volume injection of hypertonic saline can dramatically improve circulatory function during hemorrhagic shock, as evidenced by expansion of plasma volume, increased cardiac output, and reduced peripheral resistance.

Key words: hemorrhage, hypertonic saline, hypotension, hypovolemia, osmolarity, resuscitation, sheep, shock

INTRODUCTION

Infusion of large volumes of isotonic salt solution is a highly successful and universally used initial treatment of hemorrhagic shock Hyperosmolar solutions may be equally effective as a resuscitative fluid in experimental animals [1–4] and man [5–7]. Although both isotonic and hyperosmolar solutions can effectively restore

Submitted for publication September 3, 1983.

Address reprint requests to Dr. G.C. Kramer, Department of Human Physiology, University of California, Davis, CA 95616.

vascular volume in shock, hyperosmolar resuscitation may have additional beneficial effects on the cardiovascular system. Hypertonic saline infusion increases myocardial contractility [1,8] and directly vasodilates the precapillary resistance vessels [9]. This vasodilatory effect apparently explains the reduced total peripheral resistance and associated increased mesenteric blood flow and oxygen uptake reported with hypertonic resuscitation [1, 8, 10].

Our laboratory has previously reported on a comparison of isotonic lactated Ringer's and hypertonic saline (600 mOsm/liter) infusions during hypovolemic shock in unanesthetized adult sheep [11]. We found that approximately 3 liters of either solution were required to return central venous pressure to its preshock value. The hypertonic resuscitation resulted in a significantly increased cardiac output coupled with reduced pulmonary vascular resistance.

A different approach to hyperosmolar resuscitation has been taken by Velasco et al [10], who found small volumes of very hypertonic saline (2,400 mOsm/liter) could successfully resuscitate anesthetized dogs from severe hemorrhagic shock. They reported that intravenous infusions of hypertonic saline equal in volume to only 10% of shed blood caused an immediate restoration of cardiac output, arterial pressure, and visceral blood flow. This improved circulatory function occurred without an increase in measured vascular volume and was sustained for 12 h without additional fluid therapy. From both a theoretical and practical viewpoint, these observations merit further examination.

The present report examines the effects of resuscitation with small volumes of either hypertonic saline (2,400 mOsm/liter) or normal saline in hypovolemic unanesthetized sheep. In each animal we produced two episodes of shock several days apart in order to effectively compare the two resuscitation regimens. It was our goal to determine if the dramatic and sustained effects reported in anesthetized dogs could be duplicated in unanesthetized sheep subjected to moderate hemorrhagic shock.

METHODS

Animal Preparations

Six adult female sheep, 40–50 kg, were anesthetized with halothane/nitrous oxide and surgically prepared with chronic cannulations of the thoracic aorta and superior vena cava using silastic catheters placed through a neck incision. A Swan-Ganz thermodilution catheter was placed to allow monitoring of central venous pressure, pulmonary artery pressure and cardiac output. Animals were kept unrestrained in cages and had free access to food and water until 36 h before an experiment, when both food and water were removed. The first experiment was performed 3–5 days after surgery. The day of an experiment a Foley catheter was inserted into the bladder for urine collection.

Measurements

Aortic, central venous, and pulmonary artery pressures were measured with Gould P23 Db pressure transducers and continuously recorded on a multichannel strip chart recorder. Transducers were leveled to the point of the shoulder. Cardiac output was measured using an Edwards Cardiac Output Computer. Blood gases were measured with an Instrumentation Laboratories Blood Gas Analyzer. Urine was collected in a closed drainage bag; volume was determined every 30 min using a graduated

cyclinder. Hematocrit was measured and plasma protein was determined with Biuret assay on arterial blood samples taken every 30 min. Serum sodium and potassium were measured on a Nova I Na^+/K^+ Analyzer. Plasma volume was measured with the dye dilution technique [12]. Intravenous injection of 10–20 mg Evans Blue (Harvey Labs) was followed by the sampling of arterial blood after 10, 20, and 30 min. Dye concentration in plasma was measured in a Gilford spectrophotometer. Regression analysis was used to determine dye concentration at time of injection. Plasma volume was calculated as dose injected divided by plasma concentration at time of injection. Total peripheral resistance (TPR, dyne·sec/cm^5) was calculated as the difference of mean arterial and central venous pressures (mm Hg); then this quantity was multiplied by 80 and divided by cardiac output (liter·min^{-1}). Minute work (N·m·min^{-1}) was calculated as mean arterial pressure multiplied by cardiac output multiplied by 1.33×10^{-4}.

Experimental Protocol

Experiments were performed on unanesthetized animals kept unrestrained in cages. After a 2–3-h control period of data collection, the sheep were bled to a mean arterial pressure (MAP) of 50 mm Hg through their venous canulae. All shed blood was stored in standard ACD blood bags (Fenwal Laboratories). The MAP was then maintained for 2 h at 50–60 mm Hg by further bleeding as required.

At the end of the 2-h shock period, animals were resuscitated by a bolus injection of either hypertonic saline, 2,400 mOsm/liter (HS), or normal saline (NS) and observed during the next 2 h. The response after injection was monitored for 2 h. The injected volumes of each resuscitative fluid was equal to 10% of total shed blood volume. Injections were given over 60 s and no subsequent resuscitative infusion followed.

After the 2-h postresuscitation period, all shed blood was returned. All animals recovered and then underwent a subsequent shock experiment 4–7 d after the first experiment using the same protocol except with the alternate solution. The identities of the resuscitative solutions were single blind coded and the sequence of solutions was determined with random-number tables.

Plasma volumes were measured during the preshock period, after 90 min of hypotension, and 10 min after resuscitation.

Statistical Analysis

Averaged values are expressed as mean ± standard error. One-way analysis of variance was used to determine if a variable changed with respect to time after resuscitation [13]. The paired Student t test was used to compare variable differences between the two solutions used on each sheep. Differences were considered significant when $P < .05$.

RESULTS

Mean hemodynamic and blood variables are shown in Tables I and II. Measured plasma volume during the baseline period was 1,853 ± 109 ml for the 12 experiments. Assuming an F cell ratio equal to 1.0 [14] we calculated baseline blood volume to be 2,657 ± 157 ml or 56.5 ml/kg.

TABLE I. Hemodynamic Changes During Hemorrhage and Small-Volume Resuscitation†

	Baseline period	Shock period (min)		Postresuscitation (min)			
		60	120	15	30	60	120
Normal saline treatment							
MAP*	108.0 ± 16.0	58.3 ± 5.7	57.8 ± 7.7	83.3 ± 13.8	76.3 ± 12.9	76.0 ± 14.0	72.0 ± 9.8
CVP	0.4 ± 0.6	−5.3 ± 1.0	−6.3 ± 1.0	−4.4 ± 1.0	−5.4 ± 1.0	−5.8 ± 1.0	−4.4 ± 0.9
HR	101.0 ± 12.8	88.0 ± 4.1	118.0 ± 14.1	153.0 ± 16.3	158.0 ± 22.0	147.0 ± 15.8	140.0 ± 13.7
Hypertonic saline treatment							
MAP*	123.3 ± 17.4	53.3 ± 4.5	53.3 ± 6.4	100.8 ± 15.0	107.5 ± 15.1**	85.0 ± 14.9	95.0 ± 14.3
CVP	0.6 ± 0.8	−3.3 ± 1.1	−5.5 ± 1.7	−1.5 ± 1.0**	−2.4 ± 1.0	———	−4.2 ± 0.8
HR	77.9 ± 11.2	83.3 ± 5.1	127.0 ± 19.3	182.0 ± 18.9	192.0 ± 13.8**	186.0 ± 12.4	177.0 ± 14.8

†*MAP*, mean arterial pressure in mm Hg; CVP, central venous pressure in mm Hg; *HR*, heart rate.
**P* < .05 difference after injection, analysis of variance.
***P* < .05 HS compared with NS, paired Student t test.

TABLE II. Blood Variables During Hemorrhage and Small-Volume Resuscitation†

		Baseline period	Shock period (min)		Post resuscitation (min)		
			60	120	30	60	120
Normal saline							
Hct	%	29.2 ± 1.2	27.3 ± 1.2	25.1 ± 2.0	24.3 ± 2.7	26.6 ± 2.9	26.3 ± 2.9
[P]	g/100 ml	7.7 ± 0.4	6.3 ± 0.3	5.3 ± 0.3	5.2 ± 0.3	5.1 ± 0.3	5.4 ± 0.4
Na+	mEq/liter	145.8 ± 1.4	143.4 ± 1.6	143.6 ± 1.8	144.1 ± 2.3	143.4 ± 2.1	143.1 ± 2.3
K+	mEq/liter	3.9 ± 0.1	4.2 ± 0.3	4.1 ± 0.2	3.9 ± 0.4	3.7 ± 0.3	3.6 ± 0.2
Hypertonic saline							
Hct	%	31.1 ± 1.1	26.7 ± 1.1	26.6 ± 1.3	23.3 ± 1.1	24.1 ± 1.2	23.8 ± 1.5
[P]	g/100 ml	7.4 ± 0.4	6.1 ± 0.4	5.6 ± 0.4	4.9 ± 0.3	5.2 ± 0.3	5.2 ± 0.3
Na+*	mEq/liter	144.3 ± 1.2	142.3 ± 1.5	143.9 ± 1.5	153.5 ± 1.9**	151.0 ± 2.0**	151.0 ± 1.5**
K+*	mEq/liter	3.9 ± 0.1	4.3 ± 0.2	3.7 ± 0.1	2.8 ± 0.2**	3.1 ± 0.1	3.3 ± 0.1

†[P], plasma protein concentration, g/100ml.
**P* < .05 difference after injection, analysis of variance.
***P* < .05 HS compared with NS, paired Student t test.

Shock Period

No significant differences were found between the two paired experiments during the shock period. Mean arterial blood pressure was lowered to 50 mm Hg within 30 min with an initial removal of 1,100–1,300 ml of blood. After blood pressure fell below 60–70 mm Hg each animal would lie down in its cage. The animals experienced no apparent pain during the hypotension and were generally lethargic but conscious for the entire shock period. Although arterial pO_2 did not deviate from its baseline levels (85–95 mm Hg), respiratory rate invariably increased three to four times during hypotension.

Maintenance of blood pressure at 50–60 mm Hg required the removal of an additional 300–500 ml blood over the last 90 min of the shock period. Average shed blood volume was 35 ml/kg or 62% of initial blood volume. Plasma volume measured at 90 min into the shock period was 1,520 ml (Fig. 1); calculated blood volume was 2,051 ml. Comparison of preshock and shock blood volumes with shed blood indicates that over 60% of the volume of blood loss had been replaced by an autotransfusion of extravascular fluid and cells.

During the shock period central venous pressure remained 5–7 mm Hg below baseline (Table I); pulmonary artery pressure decreased in all animals from a mean baseline value of 15.0 to 10.8 mm Hg. After blood pressure had been reduced to 50 mm Hg, cardiac output decreased to 40–50% of baseline and remained at this level during the remainder of the shock period (Fig. 2).

Resuscitation

After each animal received a bolus injection of either hypertonic saline (HS), or normal saline (NS), there was an immediate increase in mean arterial pressure (Table I). Within 2 min after injection of HS arterial pressure increased 48 mm Hg and mean pulse pressure increased to 60 mm Hg. After NS the mean arterial pressure was

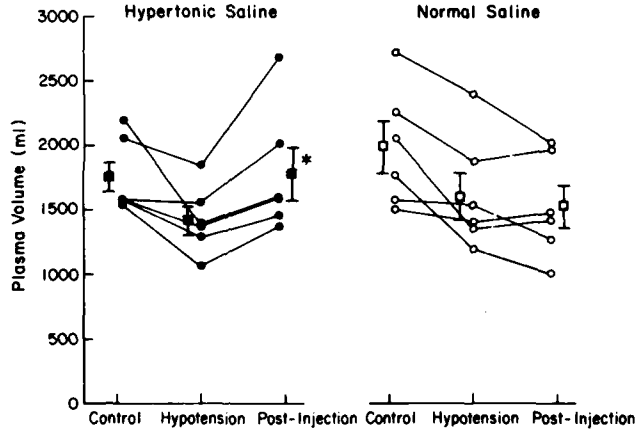


Fig. 1. Changes of plasma volume during shock and after resuscitation in both groups are shown. Plasma volume was significantly increased after hypertonic saline to a level similar to its baseline value. After normal saline, mean plasma volume remained similar to its shock value. $*P < .05$, plasma volume increased after hypertonic saline.

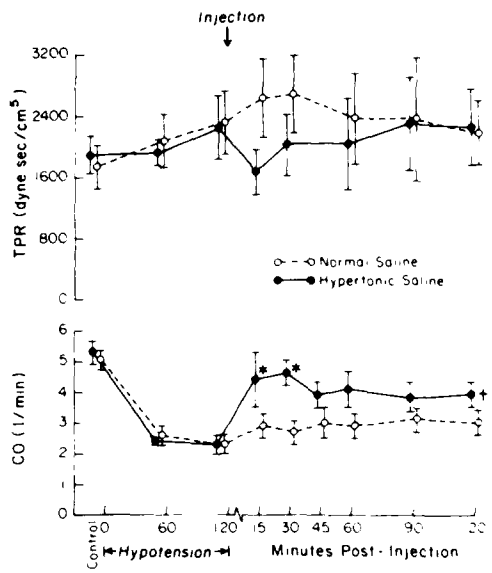


Fig. 2. Effects of hemorrhagic shock and resuscitation on cardiac output (CO) and total peripheral resistance (TPR) are shown. CO was increased significantly at 15 and 30 min after injection of hypertonic saline. TPR reduced transiently after administration of hypertonic saline. TPR after the injection of normal saline continued to increase. Means ± 1 SE. *$P < .05$, hypertonic saline compared with normal saline. †$P < .05$, increased CO after hypertonic saline injection.

increased only 26 mm Hg and the average pulse pressure was equal to 28 mm Hg, which was significantly smaller than with HS.

Cardiac output also rapidly increased after injection of HS (Fig. 2). At 15 and 30 min post HS injection cardiac output was 88–96% of its baseline value compared to 49–58% in the NS experiments. Total peripheral resistance was reduced after HS injection, while with NS mean peripheral resistance continued to increase (Fig. 2). However, during the second hour after injection neither cardiac output nor total peripheral resistance was significantly different between the two protocols.

Stroke volume did not change significantly with injection of HS (Fig. 3). Minute work was significantly increased after injection of HS (Fig. 3).

Plasma volume measured 10 min after injection was significantly increased after HS to a level similar to its baseline value; after NS plasma volume was less than during the shock phase (Fig. 1). The mean injection volume of HS was 162 ml and caused an increase in average plasma volume of 362 ml compared to the shock period. After injection of 168 ml of NS the measured plasma volume was unchanged from its shock value. Plasma protein concentrations [P] (Table II) were similar with both protocols at all times.

Plasma concentrations of sodium and potassium were unaffected by shock and injection of NS (Table II). On the other hand, injection of HS increased plasma sodium 7% to 154 mEq/liter and decreased potassium from 3.7 to 2.8 mEq/liter.

Urinary output fell to 17–21% of baseline during the second hour of shock but was increased after injection with both protocols (Fig. 4). The HS resulted in a

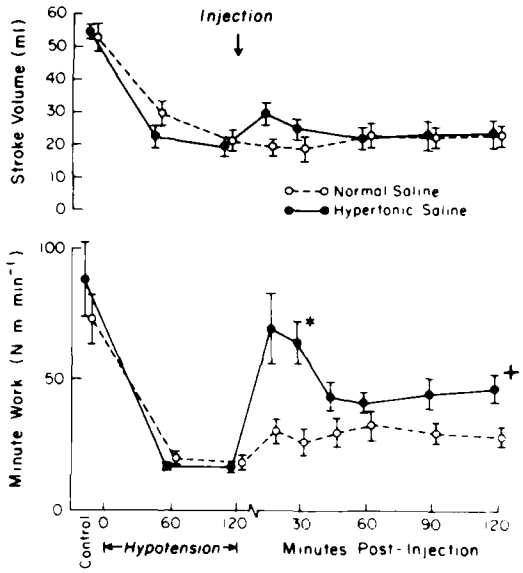


Fig. 3. Effects of hypotension and resuscitation on stroke volumes and cardiac minute work are shown. Minute work ($N \cdot m \cdot min^{-1}$) was calculated as MAP × CO × 1.33 × 10^{-4}. There was no apparent change in stroke volume after hypertonic saline injection. *$P < .05$, hypertonic saline compared with normal saline; †$P < .05$, increased cardiac work after hypertonic saline.

comparatively large output, three times baseline, during the first hour postinjection. During the same hour urine output after NS injection was less than half baseline.

DISCUSSION

We have found that the chronically catheterized unanesthetized sheep is an excellent preparation in which to study hypovolemic shock and fluid resuscitation. The level of shock induced was clinically relevant: a blood loss of more than 60% of initial vascular volume, a fall in arterial pressure to 50–60 mm Hg, and a cardiac output of less than half baseline. This moderate level of shock was completely reversible in the unanesthetized animal and allowed us to compare two resuscitation regimens during separate hypovolemic periods in each animal. The physiological responses to the two shock periods separated by a period of 4–7 d were virtually identical.

In preliminary experiments we found that a similar shock protocol in anesthetized sheep invariably resulted in death within 2 h. The striking difference in survival time between anesthetized and unanesthetized animals underlines the effectiveness of the physiological reserves that the conscious animal can initiate.

Our objective was to determine the effectiveness of small-volume infusion of very hypertonic saline in the initial resuscitation of hypovolemic shock. A recent study [10] suggested that infusion of a small volume of hypertonic saline (2,400 mOsm/liter) can permanently reestablish normal cardiovascular function during se-

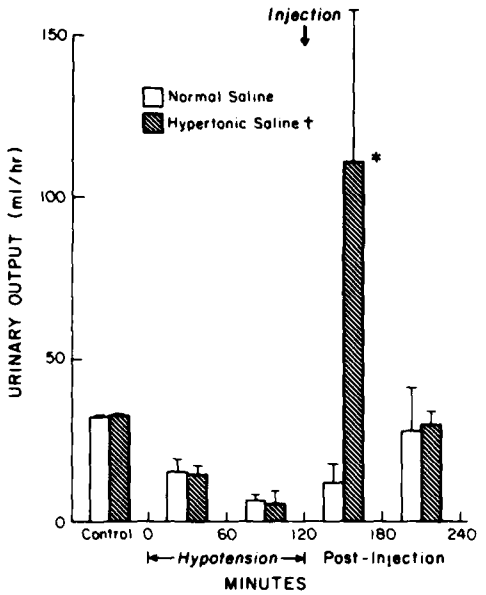


Fig. 4. Effects of resuscitation on urinary output are shown. Urinary output after hypertonic saline is markedly increased. During the first hour after injection of hypertonic saline urine flow rate was three times baseline. $^{*}P < .05$, hypertonic saline compared with normal saline; $^{\dagger}P < .05$, increased urinary output after hypertonic saline.

vere hemorrhagic shock. Other studies indicate that hypertonic saline can effectively be used for volume replacement, but the observed beneficial effects were described as transitory [8,15]. Specifically we wanted to monitor cardiovascular dynamics after small-volume resuscitation and determine if the observed beneficial effects were sustained.

Shock Period

Significant physiological reserve was demonstrated by unanesthetized sheep in response to removal of an average of 1.6 liters of blood. Ninety minutes into the shock period 69.8% of the removed plasma and 45.6% of the red cell deficit had been replaced by autotransfusion. This replenishment of vascular volume probably resulted from a combination of the following factors: the sheep's contracting spleen [16], capillary refill due to a lowered capillary pressure [17], a glucose-induced hyperosmolality [4, 18], and increased peripheral lymph flow [11, 19]. The extensive autotransfusion necessitated frequent withdrawal of blood during the entire shock period to prevent a significant return of blood pressure and cardiac output. In preliminary experiments we found that in order to induce a reproducible level of shock, animals had to be deprived of water for 36 h before each experiment.

Fluid Resuscitation

In a previous investigation [11] we reported that an average of 3.2 liters of isotonic lactated Ringer was required to return and maintain central venous pressure in adult sheep after 2 h of hypovolemic shock. This amount of fluid, slightly over twice the volume of shed blood, is representative of volumes used in current clinical resuscitation regimens. In the present study, we examined the effects of resuscitation with a fluid volume equal to one twentieth the volume used in our previous studies. Injection of 168 ml of normal saline significantly increased arterial pressure from 57.8 to 83.3 mm Hg, but small changes were observed in cardiac output or central venous pressure. By contrast, the most apparent effect of hypertonic saline was the increased cardiac output, which returned to 96% of its preshock value. Hypertonic saline injection also caused significant increases in both arterial pressure and central venous pressure, and caused a fall in total peripheral resistance.

Undoubtedly, some of these changes are due to the extracellular volume expansion caused by infusion of an osmotic load. We can estimate normal intracellular water = 40% of body weight, extracellular water = 20%, hematocrit = 30%, and osmolality = 300 mOsm/liter. After a blood loss of 1,600 ml in a 45-kg sheep and infusion of 160 ml of hypertonic saline, we calculate that ~710 ml of water moves from cells into the extracellular space. Assuming that the 870 ml expansion (160 + 710) is distributed between the blood and interstitium in a 1:2 ratio we predict an expansion of plasma volume of ~300 ml. This is in reasonable agreement with the measured increase in average plasma volume of 360 ml. Plasma volume generally continued to decline with normal saline. The average difference in plasma volume expansion between the two protocols was 255 ± 118 ml.

The decrease in cellular water, estimated to be about 4%, did not cause any observed deleterious effects. Obviously, there is a limit to the amount of hypertonic saline that can be safely infused, but 3–4 ml/kg appears safe based on our studies in sheep and the studies of Velasco et al in dogs [10]. The rise in serum sodium to 153 mEq/liter is not as alarming as the observed fall in serum potassium from 3.7 to 2.8 mEq/liter (Table II). This rapid 24% decrease in potassium concentration cannot be explained by expansion of the extracellular space, estimated to be = 10%. Neither can it be explained by renal loss, which is insignificant at 10 min after injection. Further research is required to define the mechanisms and dangers of electrolyte changes subsequent to resuscitation with very hypertonic saline.

Another clear difference between the two protocols was the fall in total peripheral resistance after hypertonic saline, while peripheral resistance remained unchanged with normal saline. Although this decrease in total peripheral resistance results partly from the expanded vascular volume, direct vasodilatory action of hyperosmolality may also contribute. Direct vasodilatory effects of hypertonicity have been well documented [9, 20] and may help explain the increased cardiac output in our study as well as improved peripheral blood flow reported in other studies [8, 11].

Hypertonicity may directly improve cardiac performance by increasing contractility and cardiac efficiency [1, 8, 21–23]. We found that hypertonic saline increased heart rate when compared to normal saline despite a higher arterial pressure with hypertonic saline. Baroreceptor feedback should be greater in the hypertonic group because of the higher arterial pressure; thus the greater heart rate is not easily

explained. A direct chronotropic effect may be beneficial but in a compromised myocardium the increased oxygen consumption could be deleterious. Minute work [24] was significantly increased through the first 30 min after hypertonic infusion. Heart rate and cardiac output increased the same relative amounts as there was no apparent change in stroke volume (Fig. 3). This contrasts somewhat with other studies in which stroke volume was increased [1, 10] after hypertonic saline infusion.

Another consistently observed effect of hypertonic saline was diuresis: Urine output was at least three times baseline during the first hour after HS injection. Glomerular filtration rate may have increased secondary to improved renal perfusion, but we have no direct evidence to support this. This apparently inappropriate diuresis during hypovolemia was surprising since hypertonic saline infusions have been shown to cause release of anti-diuretic hormone [25]. Hypertonic saline treatment could prove advantageous if it increases renal blood flow and urinary output during shock-induced renal insufficiency.

Although our study clearly showed that a small volume injection of hypertonic saline caused rapid and significant improvement in cardiovascular function, the improvement was not permanent. Arterial pressure and cardiac output both decreased from their early post injection levels. During the second hour after injection there was not a statistically significant difference between the two solutions for either cardiac output or arterial pressure although averaged values remained greater in the hypertonic saline group.

Despite the lack of a permanent effect, small-volume resuscitation could offer important advantages as an initial therapy. Rapid mobilization of cellular water and other direct cardiac and vascular effects of hypertonicity might return cardiovascular function more rapidly than traditional resuscitation. Thus, hypertonic saline treatment could prove beneficial as the initial treatment of shock and could be followed by a larger volume of isotonic fluid as needed.

ACKNOWLEDGMENTS

We wish to thank Ted Wandzilak for his excellent technical assistance and Mrs. Judy Hendrickson and Mrs. Phoebe Ling for preparing the manuscript. This work was supported by Department of Defense Contract No. DAMD 17-83-C-3175. Dr. Nakayama is supported by a fellowship from the Ministry of Health and Welfare, Japan. Dr. Kramer is a pulmonary fellow of the Francis B. Parker Foundation.

REFERENCES

1. Rowe GG, McKenna DH, Corliss RJ, Sialer S: Hemodynamic effects of hypertonic sodium chloride. J Appl Physiol 32:182–184, 1972.
2. Moylan Jr. JA, Recker JM, Mason AD: Resuscitation with hypertonic lactated saline in thermal injury. Am J Surg 125:580–584, 1973.
3. Brooks DK, Williams WG, Manley RW, Whiteman P: Osmolar and electrolyte changes in hemorrhagic shock, hypertonic solutions in the prevention of tissue damage. Lancet 521–527, 1963.
4. McNamara JJ, Mills D, Aaby GV: Effect of hypertonic glucose on hemorrhagic shock in rabbits. Ann Thorac Surg 9:116–121, 1970.
5. Monafo WW, Chuntrasakul C, Ayvazian VH: Hypertonic sodium solutions in the treatment of burn shock. Am J Surg 126:778–783, 1973.
6. Shimazaki S, Yoshioka T, Tanaka N, Sugimoto T, Onji Y: Body fluid changes during hypertonic lactated saline solution therapy for burn shock. J Trauma 17:38–43, 1977.

7. De Felippe J, Timoner J, Velasco IT, Lopes OU, and Rocha-e-Silva M: Treatment of refractory hypovolemic shock by 7.5% sodium chloride injections. Lancet 2:1002–1004, 1980.
8. Baue AE, Tragus ET, Parkins WM: A comparison of isotonic and hypertonic solutions and blood on blood flow and oxygen consumption in the initial treatment of hemorrhagic shock. J Trauma 7:743–756, 1967.
9. Gazitua S, Scott JB, Chou CC, Haddy FJ: Effect of osmolarity on canine renal vascular resistance. Am J Physiol 217:1216–1223, 1969.
10. Velasco IT, Pontieri V, Rocha-e-Silva M Jr, Lopes OU: Hyperosmotic NaCl and severe hemorrhagic shock. Am J Physiol 239:H664–H673, 1980.
11. Nerlich M, Gunther R, Demling RH: Resuscitation from hemorrhagic shock with hypertonic saline or lactated ringer's (Effect on the pulmonary and systemic microcirculations). Circ Shock 10:179–188, 1983.
12. Gibson JG, Evans WA Jr: Clinical studies of the blood volume. I. Clinical application of a method employing the azo dye 'Evans Blue' and the spectrophotometer. J Clin Invest 16:301–316, 1937.
13. Wallenstein S, Zucker CL, Fleiss JL: Some statistical methods useful in circulation research. Circ Res 47:1–9, 1980.
14. Blunt MH: "The Blood of Sheep, Composition and Function." New York: Springer-Verlag, 1975, p 7.
15. Wolf MB: Plasma volume dynamics after hypertonic fluid infusing in nephrectomized dog. Am J Physiol 221:1392–1395, 1971.
16. Hodgetts VE: The dynamic red cell storage function of the spleen in sheep. Aust J Exp Biol 39: 187–196, 1961.
17. Starling EH: On the absorption of fluids from the connective tissue space. J Physiol 19:312–326, 1896.
18. Friedman SG, Pearce FJ, Drucker WR: The role of blood glucose in defense of plasma volume during hemorrhage. J Trauma 22:86–91, 1982.
19. Drucker WR, Chadwick CDJ, Gann DS: Transcapillary refill in hemorrhage and shock. Arch Surg 116:1344–1353,1981.
20. Lundvall J, Mellander S, White T: Hyperosmolarity and vasodilation in human skeletal muscle. Acta Physiol Scand 77:224–233, 1969.
21. Koch-Weser J: Influence of osmolarity of perfusate on contractility of mammalian myocardium. Am J Physiol 204:957–962, 1963.
22. Stainsby WN, Barclay JK: Effect of infusions of osmotically active substances on muscle blood flow and systemic blood pressure. Circ Res 28 (Suppl I):I-33–I-38, 1971.
23. Wildenthal K, Mierzwiak DS, Mitchell JH: Acute effects of increased serum osmolarity on left ventricular performance. Am J Physiol 216:898–904, 1969.
24. Katz AM: "Physiology of Heart." New York: Raven Press, 1977, pp 213–214.
25. Hatzinikolaou P, Gavras H, Brenner HR, Gavras I: Sodium induced elevation of blood pressure in the anephric state. Science 209:935–936, 1980.

In conducting the research described in this report the investigators adhered to the NIH guidelines for the use of experimental animals.

Appendix
#2

JOURNAL OF SURGICAL RESEARCH 38, 180–186 (1985)

Infusion of Very Hypertonic Saline to Bled Rats: Membrane Potentials and Fluid Shifts[1]

SHIN-ICHI NAKAYAMA, M.D., GEORGE C. KRAMER, PH.D., RICHARD C. CARLSEN, PH.D., AND JAMES W. HOLCROFT, M.D.

Departments of Surgery and Human Physiology, University of California, Davis, Medical Center, Sacramento, California 95817

Submitted for publication November 10, 1983

Anesthetized rats were subjected to a moderate degree of hemorrhagic shock, lowering their mean arterial pressure to approximately 50 mm Hg for approximately 100 min. At the end of the shock period, resting skeletal muscle transmembrane potentials had depolarized from a baseline value of −82 mV to −65 mV; intracellular water had increased by 13%; and intracellular sodium and chloride contents had doubled. Eight rats were then given an infusion of very hypertonic saline (2400 mOsmole/kg, calculated osmolality) in a volume equal to only 10% of the volume of shed blood; another eight rats were given the equivalent amount of sodium and chloride in an isotonic solution (volume equal to 80% of shed blood). The mean arterial pressure in the rats that were given the very hypertonic saline returned to 81 mm Hg, compared to 55 mm Hg in the animals given normal saline. The membrane potentials in the hypertonic group polarized back to near normal— −78 mv— compared to no changes in the normal saline group. Intracellular water returned to preshock values in the hypertonic group as did intracellular sodium and chloride contents. Cellular contents in the normal saline group remained at shock levels. It was concluded that, in rats, infusion of small amounts of hypertonic saline can reverse some of the cellular abnormalities induced by hemorrhagic shock.

INTRODUCTION

Water shifts into the cells of skeletal muscle during hemorrhagic shock [5]. Administration of hypertonic saline [1, 4, 6] might be expected to draw some of this water out of these swollen cells. We examined the effects of administering very small quantities of very hypertonic saline (2400 mOsmole/kg) on skeletal muscle water in anethetized rats that had been subjected to hemorrhagic shock. We also examined the effects of the hypertonic saline on intracellular sodium and chloride contents and on skeletal muscle resting transmembrane potentials. These effects were compared with those induced by infusing an equivalent amount of solute administered as normal saline.

[1] Presented at the Annual Meeting of the Association for Academic Surgery, Syracuse, New York, November 2–5, 1983.

METHODS

Wistar rats (257 to 484 g) were allowed free access to food and water until 1 hr before being anesthetized with chloralose–urethane (3–4%, 2 ml/kg, intraperitoneally). They were then restrained in the supine position on a heating pad that was thermostatically controlled to maintain a rectal temperature of 37°C. The right femoral artery and vein were cannulated with polyethylene tubing, the ends of the catheters being placed approximately in the abdominal aorta and inferior vena cava. Heart rate was recorded and blood pressure was monitored with a pressure transducer. Resting skeletal muscle transmembrane potentials were measured with a microelectrode in individual cells of the femoral muscles after exposing the muscles by incising the overlying skin. A 30-mg piece of biceps or pectoralis major muscle was excised for determination of muscle water

0022-4804/85 $1.50

and electrolytes. One-half milliliter of blood was drawn into a syringe that was coated with sodium heparin for measurement of hematocrit and plasma sodium, potassium, and chloride concentrations.

Blood was then withdrawn out of the venous catheter until the animal's mean arterial pressure fell to 50 mm Hg, the hemorrhage lasting approximately 10 min. The shed blood was anticoagulated with sodium heparin and stored at room temperature. Arterial blood pressure was maintained at 50 mm Hg for 90 min by further hemorrhage as required. At the end of this shock period, membrane potentials were measured, a muscle biopsy was obtained, and blood was drawn for hematocrit and electrolyte determinations, the blood being replaced volume for volume by the previously shed blood. These procedures took approximately 15 min.

The animals were then entered into one of three groups: (1) eight animals were resuscitated with very small quantities (volume = 10% of shed blood) of very hypertonic saline (2400 mOsmole/kg, calculated osmolality); (2) eight animals were resuscitated with larger quantities (volume = 80% of shed blood) of normal saline (308 mOsmole/kg, calculated osmolality); (3) six animals were followed without resuscitation. The resuscitated fluids were infused through the femoral venous catheter. The hypertonic saline was infused over a period of 1 min; the normal saline over 2 min. No other resuscitative measures were undertaken: no blood was given, except to replace blood that was withdrawn for measurement of hematocrit and electrolytes; no maintenance fluids were given.

Thirty minutes after infusion of the hypertonic saline or normal saline, membrane potentials were measured, a muscle biopsy was obtained, and blood for hematocrit and electrolytes was withdrawn. Similar measurements were made between 60 and 90 min after infusion of the fluids.

Resting skeletal muscle potentials were measured with a Ling–Gerard microelectrode [5]. Potentials were recorded in at least 25 cells; these values were averaged to give the resting potential. Biopsies of skeletal muscle were taken from deep within the muscle mass. One part of the biopsy was dried to constant weight at 75°C to determine total water content. Two hundred microliters of Triton X detergent was added to a second part. The biopsy-detergent mixture was agitated at room temperature for 24 hr and then heated to 80°C in a water bath for 24 hr. The resultant suspension was centrifuged to produce a clear supernatant. Sodium and potassium concentrations in the supernatant, and in plasma, were determined by flame photometry; chloride concentrations were determined by a digital chloridometer. All determinations were performed in duplicate. Intracellular and extracellular water and electrolytes were calculated as described previously [2].

Results in the tables and text are expressed as means ± 1 standard deviation (SD) to describe distributions of the measurements. Results in the figures are displayed as means ± standard error of the mean (SE) to facilitate comparison of the two treatment regimens. A one-way analysis of variance over time was used to determine if infusion of a solution was associated with a significant change in a variable. When such associations were demonstrated, a two-tailed t test with the Bonferroni correction for multiple comparisons was used to test differences between the two treatment groups. Significance was accepted at the 5% level.

All experiments were conducted in accordance with institutional standards for care of laboratory animals.

RESULTS

A representative experiment in a 409-g rat is indicated in Fig. 1. During the baseline period, the mean resting skeletal muscle membrane potential was −84 mV. Just before time zero, blood and muscle were obtained for measurement of hematocrit, plasma electrolytes, muscle water, and muscle electrolytes. Starting at time zero and continuing for the next 90 min, 4.1 ml of blood was

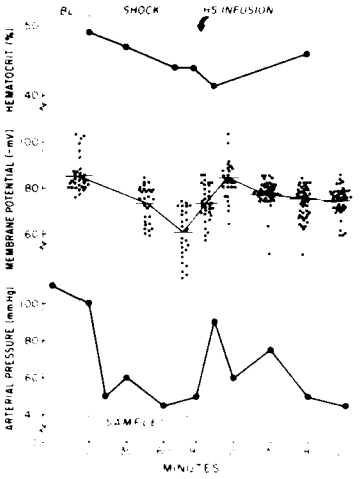


FIG. 1. Representative experiment. Each point for membrane potentials represents measurement in a single cell; horizontal lines are means of all measurements. Open vertical arrows indicate sampling of blood and muscle. BL = baseline period; HS = hypertonic saline.

withdrawn to produce a mean arterial pressure of 50 mm Hg. At 85 min the mean membrane potential had deteriorated to −60 mV. Blood and muscle were obtained. At 95 min, hypertonic saline, 2400 mOsmole/kg, 0.4 ml, was infused over 1 min. Blood pressure and potentials recovered over 30 min and then deteriorated over the next 90 min. Blood and muscle were obtained 25 and 80 min after infusion.

The animals in the three groups were subjected to the same degree of shock. The animals that were given hypertonic saline were bled slightly more than those given normal saline, but the animals in the hypertonic saline group were slightly heavier (Table 1). The animals in the normal saline group were in shock for several minutes longer than those in the hypertonic group (Table 1). By experimental design the blood pressures in the three groups at the end of the shock period were the same (Table 2). Deterioration of the membrane potentials were the same in all groups (Table 2).

The shock was moderately severe. Some animals had agonal respirations at the end of the shock period. Four of six rats given no resuscitation had died by 210 min after induction of shock (Table 1). Mean arterial pressure, 60–90 min after the end of the shock period, in the animals given normal saline or no infusion, were at shock levels (Table 2).

During shock, sodium, chloride, and water moved into the cells in all groups (Table 3). Intracellular water content, expressed as milliliters/gram dry weight of tissue, increased approximately 13%. Intracellular sodium and chloride content, at the end of the shock period, doubled compared to preshock values (Table 3). Sodium and chloride concentrations inside the cell also increased at the end of the shock period, compared to preshock values (Table 3).

Extracellular water decreased from a mean of 0.62 ± 0.14 (SD) to 0.36 ± 0.16 ml/g dry

TABLE 1

SUMMARY OF EXPERIMENTS[a]

	Hypertonic saline	Normal saline	No resuscitation
Number of animals	8	8	6
Body wt. (g)	377 ± 58	332 ± 24	355 ± 64
Shed blood (ml)	4.9 ± 1.1	3.7 ± 0.6	4.3 ± 1.0
Shock period (min)	111 ± 15	118 ± 12	—
Survival at 210 min	88% (7/8)	75% (6/8)	33% (2/6)

[a] Means ± 1 SD.

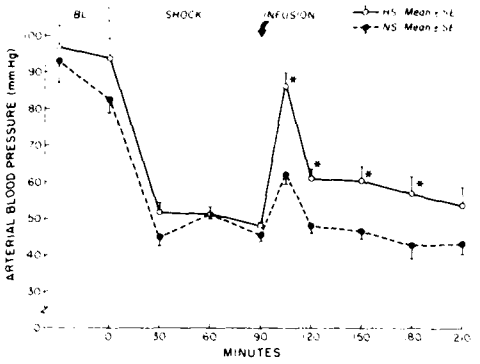


FIG. 2. Mean arterial pressures in eight rats given hypertonic saline (HS) and in eight given same total solute load as normal saline (NS). *Significant difference ($P < 0.05$) between groups.

wt during shock. Concentrations of sodium, potassium, and chloride in the plasma did not change significantly. Thus, during shock, the extracellular content of these three ions all decreased to approximately 60% of baseline values.

Infusion of hypertonic saline transiently restored all measured abnormalities back toward normal. Blood pressure came back to within 20 mm Hg of baseline values immediately after infusion and it then drifted down to shock levels (Fig. 2). Similarly, membrane potentials, after infusion of hypertonic saline, came back to almost baseline values and then gradually deteriorated (Fig. 3). Intracellular water returned to baseline values 30 min after infusion of hypertonic saline as did intracellular sodium and chloride contents (Fig. 4); these values remained close to baseline when measured 60 to 90 min after infusion (Fig. 4).

Infusion of an equivalent amount of sodium and chloride as normal saline had little effect on blood pressure (Fig. 2), membrane potentials (Fig. 3), intracellular water (Fig. 4), intracellular sodium content (Fig. 4), and intracellular chloride content (Fig. 4). These values all remained near those determined at the end of the shock period.

The differences in blood pressure, membrane potential, intracellular water, and intracellular sodium and chloride contents 30 min after infusion were all significantly different when the values in the animals given hypertonic saline were compared to the values in the animals given normal saline (Fig. 4).

DISCUSSION

The rats in all three groups were subjected to the same degree of shock. The shock was moderately severe: four of six nonresuscitated animals died within 2 hr of the end of the shock period; two of eight rats partially resuscitated with moderate amounts of normal saline died.

The hypertonic saline did not completely resuscitate the animals, but it was given as a single bolus and in extremely small quantities. Nonetheless it still rapidly corrected abnormalities of membrane potentials and of cellular sodium, chloride, and water contents, returning these values to normal within 30 min of infusion. The same amount of sodium and chloride, given as normal saline, had no beneficial effect on these cellular abnormalities.

Removal of excess cellular water by the hypertonic saline could be partly explained by hyperosmolar-induced shifts of water from the intracellular to the extracellular space. Correction of the other cellular abnormalities is more difficult to explain by the information obtained in these experiments. Even though

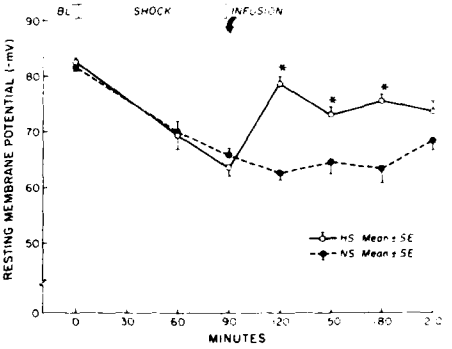


FIG. 3. Resting skeletal muscle membrane potentials. See legend for Fig. 2.

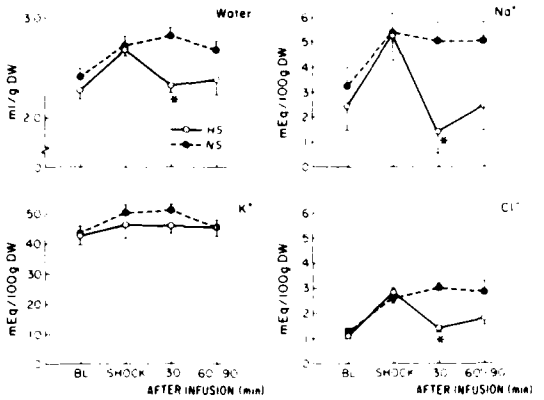


FIG. 4. Intracellular contents of water, sodium, potassium, and chloride. See legend for Fig. 2.

difficult to explain, however, the findings are striking—the skeletal muscle cells in the animals given very hypertonic saline were identical in their membrane potentials and ion contents to normal cells; skeletal muscle cells in animals given equivalent amounts of solute as normal saline and in animals given no resuscitation maintained their shock-like characteristics.

The methodology used in these experiments assumes that the Donnan equilibrium holds even in the shock state. This assumption is accepted by most workers in the field for distribution of sodium and chloride between the vascular and interstitial spaces; some workers question, however, if the Donnan equilibrium remains unchanged during shock with respect to potassium [3]. This question is impossible to answer at this time. All techniques of measuring interstitial potassium have methodologic weaknesses. Direct sampling of the interstitium by micropuncture, for example, can damage cells in the area of micropuncture; damage of only a few cells could increase interstitial potassium contents to values far higher than those actually present. In any case, the membrane potentials and the calculations of intra- and extracellular water, sodium, and chloride contents should be accurate; the calculations for intra- and extracellular potassium are the most open to question.

The weight of the animals varied, but this variation should not affect the results in any way. There are no reports in the literature that suggest that age or weight of an animal affects membrane potentials or cellular composition. In addition, during the baseline period, the membrane potentials and intracellular water, sodium, potassium, and chloride were the same in all three groups.

These studies should be taken as being strictly experimental. We do not recommend infusing very hypertonic saline to patients at this time, and we believe that much more work needs to be done on this subject before even preliminary trials in patients should be undertaken. At the same time it must be admitted that we have initiated these studies because of several features of very hypertonic saline that make it potentially attractive for use in patients. It seems to resuscitate animals rapidly, at least for 15 min or so, even when given in very small quantities, and thus might be a good solution to use in the initial resuscitation of patients subjected to trauma. The solution has a very low freezing point and is so hypertonic that bacteria cannot grow in it; thus the solutions could be stockpiled and kept for long periods of time to be used in cases of mass casualties. The solution is not viscous—in contrast to other hypertonic solutions such as glucose—and is easy to infuse. The solution also seems to have fa-

vorable hemodynamic effects, over and above those favorable effects that would be seen by plasma expansion alone.

In conclusion, the administration of very small amounts of very hypertonic saline to rats subjected to hemorrhagic shock rapidly restores membrane potentials and cellular water, sodium, and chloride contents to preshock levels. These responses are better than those seen by infusing an equivalent amount of solute in isotonic solution.

ACKNOWLEDGMENTS

We gratefully acknowledge the excellent technical assistance of Teresa Keeler and Dave Shelby and also the able secretarial assistance of Lucille Bettanini.

REFERENCES

1. Baue, A. E., Tragus, E. T., and Parkins, W. M. A comparison of isotonic and hypertonic solutions and blood on blood flow and oxygen consumption in the initial treatment of hemorrhagic shock. *J. Trauma* 7: 743, 1967.
2. Holcroft, J. W., Trunkey, D. D., and Carpenter, M. A. Extracellular calcium pool decreases during deep septic shock in the baboon. *Ann. Surg.* **192:** 683, 1980.
3. Illner, H., and Shires, G. T. The effect of hemorrhagic shock on potassium transport in skeletal muscle. *Surg. Gynecol. Obstet.* **150:** 17, 1980.
4. Lopes, O. U., Pontieri, V., Rocha e Silva, M., Jr., and Velasco, I. T. Hyperosmotic NaCl and severe hemorrhagic shock: Role of the innervated lung. *Amer. J. Physiol.* **241:** H883, 1981.
5. Shires, G. T., Cunningham, J. N., Baker, C. R. F., Reeder, S. F., Illner, H., Wagner, I. Y., and Maher, J. Alterations in cellular membrane function during hemorrhagic shock in primates. *Ann. Surg.* **176:** 288, 1972.
6. Velasco, I. T., Pontieri, V., Rocha e Silva, M., Jr., and Lopes, O. U. Hyperosmotic NaCl and severe hemorrhagic shock. *Amer. J. Physiol.* **239:** H664, 1980.

Appendix
#3

JOURNAL OF SURGICAL RESEARCH 39, 517–528 (1985)

A Comparison of Several Hypertonic Solutions for Resuscitation of Bled Sheep[1]

G. JEFFREY SMITH, M.D.,* GEORGE C. KRAMER, PH.D.,† PAUL PERRON, B.A.,† SHIN-ICHI NAKAYAMA, M.D.,* ROBERT A. GUNTHER, PH.D.,* AND JAMES W. HOLCROFT, M.D.*

*Department of Surgery, University of California, Davis Medical Center, Sacramento, California 95817, and †Department of Human Physiology, University of California, Davis, California 95616

Submitted for publication November 8, 1984

Small volumes (4 ml/kg) of 2400 mOsm NaCl restore cardiac output and mean arterial pressure to 80% of baseline after hemorrhage (65% of blood volume) in unanesthetized sheep. An equal volume of normal saline is less effective. To identify an optimal hypertonic solution, we screened six 2400 mOsm solutions in 18 randomized experiments in 8 sheep: NaCl, $NaHCO_3$, NaCl/sodium acetate, NaCl/mannitol, NaCl/6% Dextran 70, and glucose. Cardiovascular function, as determined by cardiac output and mean arterial pressure, was restored best with NaCl, NaCl/NaAc, and NaCl/Dex. These three solutions were then evaluated using 18 sheep in 36 experiments. Following a 1-hr baseline period, the sheep were bled to a mean arterial pressure of 50 mm Hg for 2 hr. One of the solutions was then given in a volume of 4 ml/kg over 2 min and the sheep were monitored for 3 hr. Within 3 min of the infusion, cardiac output increased to greater than 100% of baseline for all three solutions. The NaCl–Dex solution sustained a significantly higher cardiac output over the 3-hr observation period than the other solutions. Plasma volume increased for all solutions following infusion. NaCl–Dex maintained plasma volume significantly better than the other solutions. As a further control, an isotonic solution of 6% Dextran 70 in normal saline was studied. It was not as effective as the hypertonic NaCl–Dex in maintaining cardiac output, mean arterial pressure, or plasma volume. Osmolality increased 10% (309 to 326 mOsm/kg H_2O), plasma [NA] increased 7% (151 to 161 meq/liter), and plasma [K] decreased from 3.9 to 2.6 meq/liter following the hypertonic infusions. The sheep appeared to tolerate these electrolyte changes well. We conclude that a single bolus infusion of 2400 mOsm NaCl with 6% Dextran 70 best resuscitates sheep that have been subjected to a moderate degree of hemorrhagic shock compared to several other solutions. Its beneficial effects are caused in part by a sustained reestablishment of plasma volume. More studies are needed to document the safety of dextran in the clinical setting of hemorrhagic shock. Small volumes of hypertonic solutions may be valuable in the initial fluid resuscitation of patients in hemorrhagic shock.

INTRODUCTION

Hypertonic solutions have been used to improve cardiovascular function and tissue perfusion in a variety of clinical situations. In the 1920s, Silbert, while infusing 5% sodium chloride solutions to patients with Buerger's disease, noted a transient increase in the pulse amplitude [1]. In the 1970s, Moylan and Monafo used hypertonic solutions in burn resuscitation and found the solutions to be safe and effective [2, 3]. Recently, hypertonic sodium lactate was studied in patients undergoing elective abdominal aortic surgery; it was found to be as effective as a larger volume of isotonic solution in supporting cardiovascular function [4].

Experimentally, Velasco, in Brazil, found that small volumes of 2400 mOsm sodium chloride (7.2%) effectively resuscitated anesthetized dogs from hemorrhagic hypotension [5]. We reported similar results in unanesthetized sheep and confirmed that small volume resuscitation with hypertonic sodium chloride was effective [6, 7].

Hypertonic solutions appear to benefit cardiovascular function on multiple levels [8–10].

[1] Presented at the Annual Meeting of the Association of Academic Surgery, San Antonio, Texas, October 31–November 3, 1984.

0022-4804/85 $1.50

The pulse rate increases, precapillary arterial beds dilate, and plasma volume increases—all of these effects serving to increase cardiac output. We have also found that hypertonic saline restores both skeletal muscle membrane potential and skeletal muscle intracellular electrolytes and water after hemorrhagic hypotension [11].

The above studies did not establish whether the beneficial effects of these solutions were due to their hypertonicity per se, or to their solute composition. In this study we compared the effectiveness of six hypertonic solutions having different compositions. Our purpose was to determine the most effective constituents for a hypertonic resuscitative solution, and to compare the solutions' effectiveness based on their solute composition. The six different 2400 mOsm solutions—sodium chloride, sodium bicarbonate, sodium chloride/sodium acetate, sodium chloride/mannitol, sodium chloride/dextran, and glucose are listed in Table 1. All of these solutions have been used in the past, either clinically or experimentally, in lower concentrations, and were therefore selected for study [2, 3, 13–16]. The sodium chloride/dextran consisted of 6% Dextran 70 with 1.2 *M* sodium chloride. This solution was both hypertonic (osmolality = 2400 mOsm) and hyperoncotic (colloid osmotic pressure = 70 mm Hg) [12].

Following an initial screening, three of the solutions—sodium chloride, sodium chloride/sodium acetate, and sodium chloride/dextran—were compared in more detail. The results of the initial studies of the six solutions and of the final studies with the three solutions are reported here. Also, the results of two control studies are included—no resuscitation and dextran/normal saline resuscitation.

TABLE 1

SIX 2400 mOsm SOLUTIONS

NaCl	1.2 *M* NaCl
$NaHCO_3$	1.2 *M* $NaHCO_3$
NaCl-NaAc	0.6 *M* NaCl and 0.6 *M* Na acetate
NaCl-Man	0.7 *M* NaCl and 1.0 *M* mannitol
NaCl-Dex	1.2 *M* NaCl in 6% Dextran 70
Glu	2.4 *M* glucose

METHODS

Animal preparations. Three series of experiments were performed. In the first series of experiments, six hypertonic solutions of varying composition were screened using 8 sheep in 18 experiments (Table 1). In the second series of experiments, three of these solutions—sodium chloride, sodium chloride/sodium acetate, and sodium chloride/dextran—were compared using 16 sheep in 36 experiments. In the third series of experiments, two more control groups were added—no resuscitation following hemorrhage and resuscitation using an isotonic dextran/normal saline solution. In these experiments, each treatment was evaluated in 4 sheep in 4 experiments.

Chronic cannulation of the thoracic aorta, superior vena cava, and pulmonary artery were accomplished in female sheep, weighing 40–50 kg, under halothane/nitrous oxide anesthesia through a neck incision using silastic and Swan-Ganz thermodilution catheters. A Foley catheter was placed on the day of the experiment to monitor urine output. Food and water were withheld for 24 hr in the first series of experiments and 36 hr in the second and third series. We have found that the longer fast results in a more reproducible level of shock [6]. At least 48 hr elapsed between the intraoperative placement of the catheters and the initiation of fasting for a hemorrhage experiment.

Measurements. Mean arterial pressures and pulmonary artery pressures were measured with a Gould P23 pressure transducer connected to a multichannel strip chart recorder for continuous monitoring. Transducers were leveled to the point of the shoulder. Cardiac output was measured by thermodilution, using an Edwards Cardiac Output Computer. Urine was collected in a closed drainage system equipped with a graduated cylinder. Hematocrits were determined with a Damon IEC Microhematocrit Centrifuge. Sodium and potassium were measured by flame photometry. Blood urea nitrogen and creatinine were measured on a Gilford Clinical Chemical Analyzer System 103. Osmolality was determined on

an Advanced Instruments Incorporated Freeze Point Osmometer. Plasma volume was measured by the Evans blue dye dilution technique [17].

Experimental protocol. All experiments were conducted on unanesthetized animals kept unrestrained in cages. After an initial 1-hr period of baseline data collection, the sheep were bled to a mean arterial pressure of 50 mm Hg. The mean arterial pressure was held at 40–55 mm Hg by continued bleeding for the next 2 hr. ACD blood bags (Fenwal Laboratories) were used for blood storage.

In the first series of experiments, the sheep received a bolus infusion of 4 ml/kg of one of the six hyperosmolar solutions (Table 1), and were monitored for the next 2 hr. All blood was returned at the end of 2 hr and the experiment terminated. The mean arterial pressure, cardiac output, pulmonary artery pressures, heart rate, urine output, sodium, potassium, osmolality, hematocrit, and glucose were determined every 30 min during the experiment.

In the second series of experiments, the three best solutions—as determined by their ability to restore mean arterial pressure and cardiac output—were then studied in a blinded randomized fashion. These solutions—sodium chloride, sodium chloride/sodium acetate, and sodium chloride/dextran—were kept in stock and assigned a code number so as to prevent their identification during the course of the experiments. The solutions were given in a randomized protocol. Since the sheep could be used two or three times, the protocol prevented any solution from being used too often as the first solution. No sheep received the same solution twice. Table 2 records the results of this randomization. A 60-min baseline was followed by 2 hr of hemorrhage. A bolus infusion of 4 ml/kg was followed by a 3-hr period of observation. Plasma volume and plasma and urinary creatinine concentrations were determined in addition to the parameters mentioned earlier.

Table 3 lists the hemodynamic differences depending on whether a solution was used in a first (N = 15), second (N = 12), or third (N = 9) study. Except for hematocrit, the other parameters, while demonstrating a progressive downward trend, are similar or are not significantly different. The randomized protocol as indicated in Table 2 minimized these differences. We have previously found that our protocol of hemorrhagic hypotension is nearly always reversible with resuscitation. At the end of each experiment blood is returned and the animals recover. The response to a second or third hemorrhage several days later is essentially identical; i.e., similar falls in cardiac output and shed blood volume during the 2 hr of controlled hypotension.

TABLE 2

PROTOCOL: EVALUATION OF THREE SOLUTIONS

Solution	First	Second	Third
NaCl	5	5	3
NaCl-NaAc	5	3	3
NaCl-Dex	5	4	3

In the third series of experiments, the 2-hr hemorrhage period was followed by no resuscitation in 4 experiments in 4 sheep. Another series of 4 control experiments in 4 sheep consisted of giving 6% Dextran 70 in normal saline as a resuscitation fluid at 4 ml/kg.

Statistical analysis. Averaged values are expressed as the mean ± the standard error in the first and third series of experiments. Because of the small number of experiments for each solution, no further analysis was done. In the second series, averaged values are expressed as the mean ± the standard deviation. An analysis of variance was used to compare variable differences between the three solutions. Differences were considered significant when $P < 0.05$.

RESULTS

Mortality

Three sheep, out of a total of 32 sheep used in 62 experiments, died during the final 30 min of hemorrhage. This represented a 3–5% mortality from the shock itself. Two more

TABLE 3

HEMODYNAMIC DATA ACCORDING TO EXPERIMENT ORDER

	First	Second	Third
N	15	12	9
Hct baseline	31%	28%	25%
180 min p̄ bolus	21	19	16
MAP baseline	100 mm Hg	96 mm Hg	92 mm Hg
3 min p̄ bolus	93	93	88
180 min p̄ bolus	83	83	82
C.O. baseline	5.2 liters · min^{-1}	5.1 liters · min^{-1}	4.8 liters · min^{-1}
Hemorrhage	2.1	2.0	2.3
3 min p̄ bolus	5.6	6.0	5.6
180 min p̄ bolus	4.1	4.0	4.3
PV baseline	45 ml · kg^{-1}	43 ml · kg^{-1}	44 ml · kg^{-1}
180 min p̄ bolus	36	35	39
Blood removed	41 ml · kg^{-1}	41 ml · kg^{-1}	39 ml · kg^{-1}

sheep died after the hypertonic bolus infusion. One died immediately following infusion of a sodium chloride/sodium acetate solution; the other suffered a stroke from an air embolus flushed into the aorta from the arterial catheter 45 min following hypertonic infusion. No specific toxicity could be identified with hypertonic infusions. The single death immediately following the hypertonic infusion may have been a result of the severity of the hemorrhage as in the other 3 sheep.

First Series—Comparison of Six Solutions

Severity of shock. Eight sheep were studied in 18 experiments. There were no significant differences among the six groups during the baseline period or during hemorrhage. In the first 30 min of hemorrhage, the mean arterial pressure rapidly decreased to 50 mm Hg during which time the animals usually became lethargic and sat down in the cage. The animals became tachypnic and tachycardic during this period. The pressure was held at 50 mm Hg by continued hemorrhage. The volume of blood removed for sodium chloride, sodium bicarbonate, sodium chloride/sodium acetate, sodium chloride/mannitol, sodium chloride/dextran, and glucose was 41, 47, 41, 39, 46, and 46 ml/kg, respectively. Average was 43 ml/kg or 60–70% of the estimated blood volume.

Figures 1 and 2 show the cardiac output and mean arterial pressure for these experiments. Cardiac output was decreased to 40–45% of baseline for all solutions. During this period, heart rate and total peripheral resistance increased, and pulmonary artery pressures decreased. The serum glucose for all groups rose from a baseline average of 60–70 g% to an average of 115–125 during hemorrhage.

Response to infusion. With the infusion of 4 ml/kg of a test solution, equivalent to approximately 10% of the shed blood volume, the mean arterial pressure immediately increased to 85% of baseline for all solutions. As this bolus was delivered, the pressure began to rise. Over the 2-hr observation period, the mean arterial pressure and cardiac output gradually declined. Sodium chloride, sodium chloride/sodium acetate, and sodium chloride/dextran solutions best maintained both mean arterial pressure and cardiac output.

With the infusion of all 2400 mOsm solutions, the sheep became more alert, more active, and usually stood up. Respiratory rate decreased from its hemorrhage level of 40–60 breaths per minute to 15–20 breaths per minute, similar to baseline. Heart rate increased

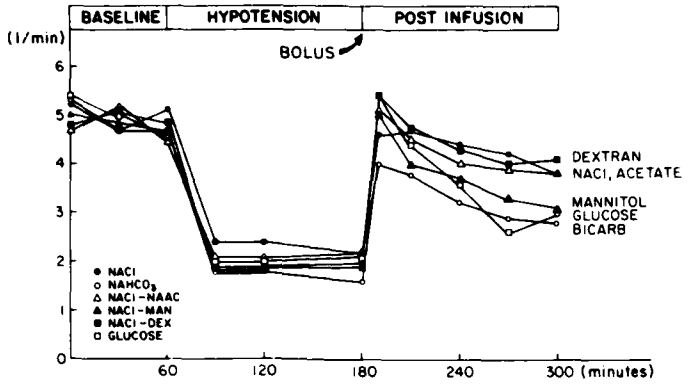


FIG. 1. The cardiac output during baseline and hemorrhage, and following hypertonic bolus is shown. While all six solutions were initially effective in restoring cardiac output, the NaCl, NaCl-NaAc, and NaCl-Dex solutions maintained this effect better than the other solutions. Mean values.

to greater than 160 beats per minute for all sheep. The effects on heart rate and respiratory rate were immediate. Total peripheral resistance increased from an average of 1.4×10^3 to 1.8×10^3 dyne·sec·cm^{-5} at the end of hemorrhage. The total peripheral resistance returned to baseline immediately after the bolus infusion with all solutions, and then gradually increased over the next 2 hr.

All solutions induced a diuresis in the first 30 min post infusion. Mannitol and glucose were associated with a diuresis of 150 and 200% of the infused volume, respectively. One animal that received a sodium bicarbonate infusion suffered a seizure. The serum pH following the seizure was 7.8. Plasma osmolality increased by 10%, ranging from 325–335 mOsm. The plasma sodium increased from 140–145 to 150–155 meq/liter following infusion of the sodium-containing solutions. Plasma potassium decreased to 2.5–3.0 meq/liter.

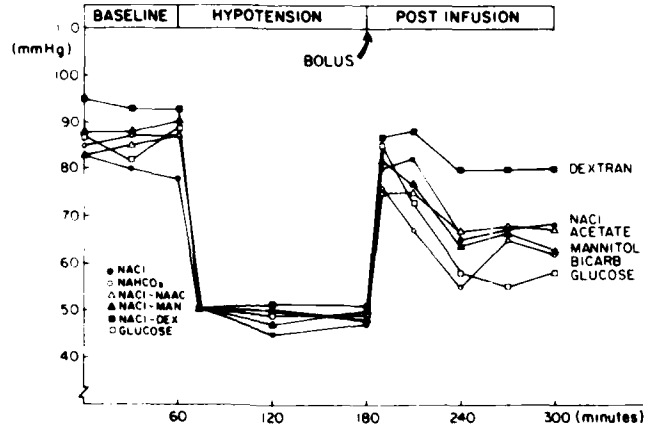


FIG. 2. The mean arterial pressure during baseline, hemorrhage, and following hypertonic bolus infusion is shown. All six solutions were initially effective. NaCl, NaCl-NaAc, and NaCl-Dex appeared to best sustain this effect. Mean values.

TABLE 4

VOLUME OF HEMORRHAGE

	N	Liters	ml · kg $^{-1}$
NaCl	13	1.72 ± 0.21	39 ± 4.1
NaCl–NaAc	11	1.91 ± 0.25	44 ± 4.8
NaCl–Dex	12	1.77 ± 0.17	40 ± 5.0

Second Series—Comparison of Three Solutions

Severity of shock. Sixteen sheep were evaluated in 36 experiments. Based on the initial screening experiments just reviewed, three solutions were compared further: sodium chloride, sodium chloride/sodium acetate, and sodium chloride/dextran. After baseline data collection, the sheep were subjected to hemorrhagic hypotension for a 2-hr period as before. A bolus infusion of 4 ml/kg of one of the solutions was given and the sheep were observed for an additional 3 hr. Table 4 lists the volumes of shed blood for the experiments with each solution.

There were no significant differences between the groups of solutions during the baseline and hemorrhage periods. As before, tachycardia, tachypnea, an increase in total peripheral resistance, and a decrease in pulmonary vascular pressures accompanied a reduction of cardiac output to 40–50% of baseline. Urine output and plasma volume also decreased as anticipated.

Response to infusion. Figures 3 and 4 demonstrate the response of cardiac output and mean arterial pressure to the bolus infusions, delivered over 1–2 min. Within 3 min of starting the infusion, the cardiac output increased to greater than 100% of baseline for all three solutions. Over the ensuing 3-hr observation period, the 2400 mOsm sodium chloride/dextran solution maintained a significantly higher cardiac output when compared to NaCl or NaCl/NaAc. The mean arterial pressure responded similarly although the differences were not significant.

Plasma volumes (Fig. 5), measured 10 min after infusion, were increased for all three solutions by at least 350 ml, or approximately 8 ml/kg. The volume infused averaged 180 ml, indicating net plasma volume expansion caused by all solutions. The immediate fall in hematocrit (Fig. 6) following infusion also reflected an expanded plasma volume. The hypertonic sodium chloride/dextran solution maintained the plasma volume significantly better than the other two solutions as indicated by the 3-hr determination.

The total peripheral resistance (Fig. 7) increased with hemorrhage and decreased immediately with the infusions. The dextran so-

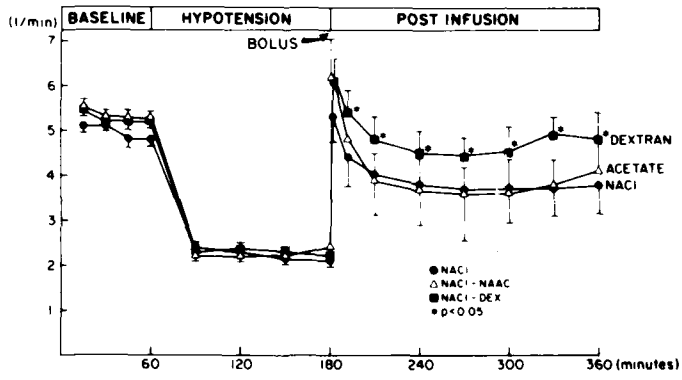


FIG. 3. The cardiac output response during the second series of experiments is shown for NaCl, NaCl–NaAc, and NaCl–Dex. All solutions initially restored cardiac output to 100% of baseline. The dextran solution maintained the cardiac output significantly higher throughout the 3-hr observation period. Mean ± SD.

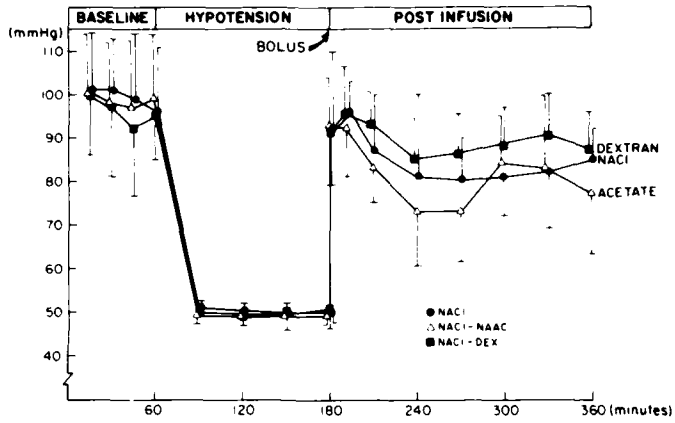


FIG. 4. The mean arterial pressure response during the second series of experiments is shown. All solutions were initially effective. NaCl–Dex maintained a higher mean arterial pressure though not significantly higher. Mean ± SD.

lution maintained a lower total peripheral resistance than the other solutions.

There were no differences in the heart rate, respiratory rate, or pulmonary artery pressures among the three solutions. The tachycardia and tachypnea associated with hemorrhage responded to the infusions with a further increase in tachycardia and a decrease in tachypnea to baseline. The pulmonary artery pressure decreased from 15 to 10 mm Hg and the wedge pressure decreased from 9 to 4 mm Hg with hemorrhage. Baseline values were restored with the infusions.

There were no significant differences between the groups for plasma sodium or potassium concentrations, plasma osmolality, blood urea nitrogen concentrations, or plasma creatinine. Table 5 lists the concentrations ± the standard deviation of these variables for the second series of experiments. The sheep were moderately dehydrated at the conclusion of a 36-hr fast and appeared to tolerate the electrolyte abnormalities induced by the hypertonic infusions well.

Urine output decreased to 20% of baseline during hemorrhage for all solutions. With the

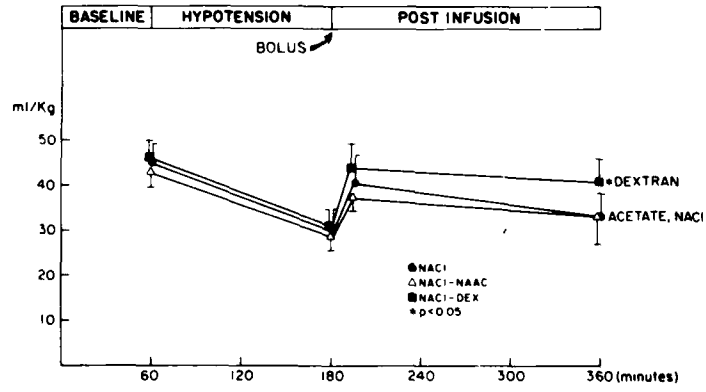


FIG. 5. Changes in plasma volume during shock and after resuscitation are shown. All solutions increased plasma volume following bolus infusion. NaCl–Dex caused a greater plasma volume expansion initially, and after 3 hours, maintained a significantly higher plasma volume than the other solutions. Mean ± SD.

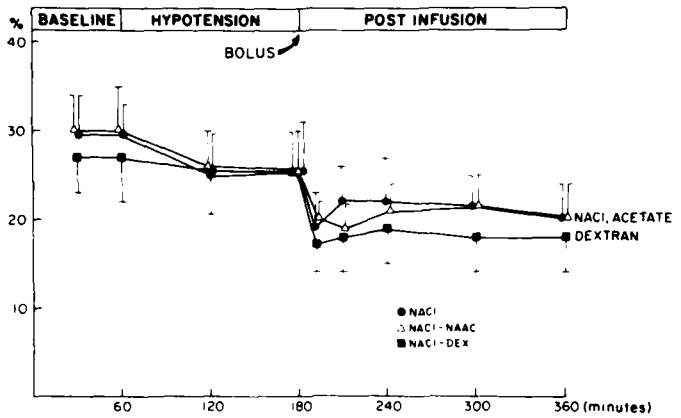


FIG. 6. Changes in hematocrit during the second series of experiments are shown. Hematocrit decreased slightly during hemorrhage. Following infusion, a large decrease was noted for all solutions, probably secondary to plasma volume expansion. Mean ± SD.

infusion of all three hypertonic solutions, the urine output increased to 100–200% over baseline during the first hour following infusion. Creatinine clearances were increased for all solutions from approximately 100 ml/min at baseline to over 200 ml/min after the bolus infusion.

Third Series—Control Experiments

Figures 8 and 9 show the cardiac output and the mean arterial pressure responses for two more control groups. The protocol for hemorrhage was identical to all of the previous experiments. The shed blood volume was 37 ml/kg for both groups. Using 4 sheep in 4 experiments in one control group, no resuscitation was given following the 2-hr hemorrhage. Cardiac output 30, 60, and 180 min after the end of hemorrhage was 2.5, 2.9, and 3.1 liters/min, respectively. Mean arterial pressure remained depressed without any bolus infusion. Plasma volume remained 30 ml/kg after the 3-hr observation period.

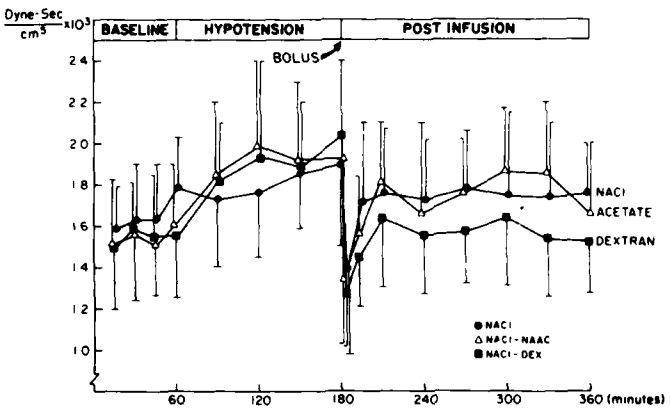


FIG. 7. The total peripheral resistance changes during hemorrhage and following infusion are shown. Resistance increased during hemorrhage and returned to baseline following hypertonic infusion. Dextran maintained a lower total peripheral resistance than the other solutions. Mean ± SD.

TABLE 5

ELECTROLYTE CHANGES WITH 2400 mOsm RESUSCITATION

			Postinfusion (min)		
	Baseline	Hypotension	10	30	180
[Na] meq/liter	151 ± 8.0	150 ± 7.0	161 ± 8.0	160 ± 5.0	159 ± 7.0
[K] meq/liter	3.9 ± 0.4	4.0 ± 0.5	2.6 ± 0.3	2.9 ± 0.3	3.0 ± 0.3
[Osm] mOsm/kg H_2O	309 ± 10	315 ± 13	334 ± 12	334 ± 11	326 ± 9
BUN mg/dl	20 ± 5.5	24 ± 5.9	24 ± 6.6	24 ± 6.9	24 ± 5.9
[CR] mg/dl	0.9 ± 0.2	1.4 ± 0.3	1.3 ± 0.3	1.3 ± 0.3	1.1 ± 0.4

In a second group of 4 experiments using 4 sheep, 6% Dextran 70 in normal saline was evaluated as a bolus infusion. Four ml/kg of the solution was infused as before and the sheep were monitored for 3 hr. The initial mean arterial pressure and cardiac output responses to the bolus infusion were not as dramatic as with the hypertonic solutions.

DISCUSSION

Velasco *et al.* rekindled interest in applying hypertonic resuscitation to hemorrhagic shock by using small volumes of 2400 mOsm sodium chloride to resuscitate anesthetized dogs following hemorrhage [5]. Our laboratory developed a hemorrhage protocol using chronically instrumented unanesthetized sheep to study hypertonic solutions in a reproducible hypovolemic state. Nakayama, using this model, demonstrated that small volumes of hypertonic saline more effectively resuscitated the sheep than a similar volume of normal saline [6].

In the first series of experiments reported here, six 2400 mOsm solutions, regardless of the solute content, rapidly restored both cardiac output and mean arterial pressure to at least 85% of baseline. Only 4 ml/kg, representing approximately 10% of the shed blood volume, was required to produce this effect. This initial improvement in cardiovascular function, represented by increases in cardiac output and mean arterial pressure seems to be mediated by the hypertonicity of the solutions; the solute composition does not seem important. Nakayama has shown that isotonic normal saline in a similar small volume does not

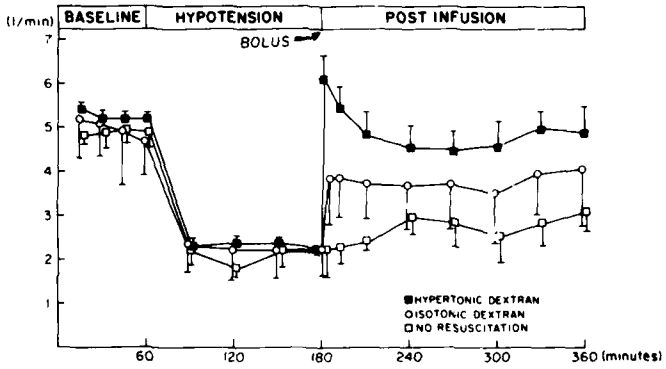


FIG. 8. The cardiac output during the third series of experiments is shown. Also shown is a no resuscitation control group. The isotonic saline/dextran saline solution did not demonstrate as large an initial increase in cardiac output as the hypertonic saline/dextran solution. Mean ± SD.

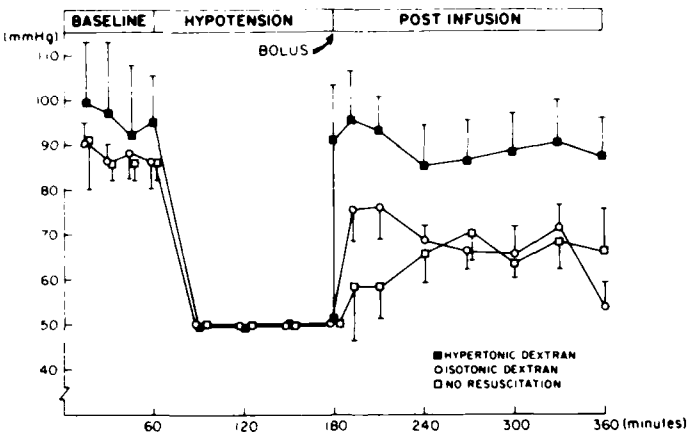


FIG. 9. The mean arterial pressure during the third series of experiments is shown. The no resuscitation control group is also shown. The isotonic saline/dextran solution was not as effective as the hypertonic saline/dextran solution in restoring or maintaining mean arterial pressure. Mean ± SD.

cause the rapid early increase in cardiac output and mean arterial pressure that is seen with all the hypertonic solutions studied here [6]. Also, in the third series of experiments, 6% Dextran 70 in normal saline, an isotonic solution, initially returned cardiac output to only 70% of baseline. The other hypertonic solutions all restored cardiac output to at least 85% of baseline immediately following infusion.

Maintenance of this favorable response was more dependent on solute than on hypertonicity. Glucose and mannitol induced an osmotic diuresis which offset the early cardiovascular benefits associated with hyperosmolality. The bicarbonate solution created an alkalosis which compromised any improvement that may have resulted from its hypertonicity. These three solutions were eliminated from further consideration.

In the second series of experiments all three solutions restored cardiac output to at least 100% of baseline within 3 min of beginning the infusion. The dextran solution, however, maintained a significantly higher cardiac output, greater than 85% of baseline, throughout the remaining 3 hr of monitoring. The hypertonic dextran solution also sustained the mean arterial pressure better than the other solutions, though this was not statistically significant. Dextran induced a greater initial plasma volume expansion and maintained the effect presumably because of its higher colloid osmotic pressure. If blood volume = [plasma volume/(1 − HCT)], then the baseline blood volumes are 64 and 63 ml/kg for the hypertonic NaCl and NaCl/dextran solution, respectively. Three hours following resuscitation the blood volumes are 42 and 50 ml/kg, respectively, an augmentation of 15–20%, indicating that the blood volume is maintained better with dextran added to hypertonic NaCl. This increased volume probably accounts for the higher cardiac output over the 3-hr observation period. Hemodilution does not explain this increase. After 3 hr, hematocrits are 20 and 18% for the saline and dextran solutions, respectively, a difference in oxygen carrying capacity of only 2–4%. The cardiac outputs at this time are 3.7 and 4.7 liters/min, respectively, a difference of approximately 25%.

The hypertonicity itself contributes some benefit to maintaining the cardiac output and mean arterial pressure following the initial infusion. This is implied in the finding that the isotonic dextran/normal saline control solution did not maintain the cardiac output and mean arterial pressure as well as the hypertonic sodium chloride/dextran solution during the 3-hr observation period. Also, increased plasma osmolality was always associated with

a decreased peripheral resistance, presumably a result of precapillary vasodilation.

Though the hypertonic dextran solution was superior to the other hypertonic formulations, dextrans may not be appropriate in the treatment of hypovolemic shock secondary to trauma. While no evidence of renal impairment was identified due to dextran in these experiments, it has been reported. Additionally, problems with the typing and crossmatching of blood would need to be addressed.

This study did not compare different levels of hypertonicities. In two preliminary experiments we infused 2 ml/kg of 4800 mOsm sodium chloride into 2 similarly hemorrhaged sheep, inducing convulsions in both before the infusions could be completed. The upper safe limits of hypertonic infusion remain to be determined although 2400 mOsm solutions appear safe.

The increases in plasma sodium and osmolality were tolerated well by the sheep despite their being somewhat dehydrated after 36 hr of absolute fasting. Also, no problems could be identified relative to the observed hypokalemia. Potassium levels remained normal in similar studies in rats [11] and in pigs [18]. All three solutions improved renal perfusion as indicated by the increased urine output and by an increase in creatinine clearance following infusion.

In our experience, the 2400 mOsm hypertonic salt solutions appear safe and effective when used as a small volume infusion. In over 100 hemorrhage experiments over the past 3 years, one animal has seized and one has died following infusion. The seizure was associated with a pH = 7.8 following infusion of hypertonic sodium bicarbonate. The death occurred following a bolus of hypertonic sodium chloride/sodium acetate and may have been caused by the degree of shock and not the solution itself.

A complete understanding of the mechanism of hypertonic resuscitation remains to be determined. However, this study established that a 4 ml/kg bolus of a hypertonic fluid increased plasma volume 8–12 ml/kg in bled sheep. This partial restoration of blood volume may be the predominant mechanism. The capillary wall is highly permeable to small solutes like sodium and chloride while the cell membrane restricts their movement. Thus, a hypertonic bolus of NaCl will osmotically move cell water from the cell into the extracellular fluid volume. We estimate that 180 ml of 2400 mOsm NaCl will pull over 700 ml out of the cellular space [6]. In shock, the extracellular fluid volume is proportioned more into the plasma volume because of low capillary hydrostatic pressure. The addition of 6% Dextran 70, which is 2.5 times more osmotically active than albumin, causes an even greater preferential redistribution of interstitial water into the vasculature.

In summary, small-volume infusion of hypertonic solutions rapidly and effectively restored cardiovascular flow. The 2400 mOsm sodium chloride, sodium chloride/sodium acetate, and sodium chloride/dextran solutions maintained these beneficial effects better than three other solutions. The hypertonic dextran solution was superior to all solutions evaluated. Rapid early restoration of organ perfusion is the goal of resuscitation to prevent multiorgan system failure. A small bolus of a hypertonic solution delivered in the field could stabilize blood pressure and cardiac output long enough to allow transportation to a treatment center. Dextran may prove beneficial in this setting although futher study is necessary.

ACKNOWLEDGMENTS

We thank Ted Wandzilak for his excellent technical assistance and Lynne Sullens for preparing the manuscript. This work was supported by Department of Defense Contract DAMD 17-83-C-3175.

REFERENCES

1. Silbert, S. The treatment of thromboangitis obliterans by intravenous injection of hypertonic salt solution: Preliminary report. *J. Amer. Med. Assoc.* **86:** 1759, 1926.
2. Moylan, J. A., Jr., Recker, J. M., and Mason, A. D. Resuscitation with hypertonic lactated saline in thermal injury. *Amer. J. Surg.* **125:** 580, 1973.
3. Monafo, W. W., Chuntrasakul, C., and Ayvazian, V. H. Hypertonic sodium solutions in the treatment of burn shock. *Amer. J. Surg.* **126:** 778, 1973.

4. Shackford, S. R., Sise, M. J., Fridlund, P. H., Rowley, W. R., Peters, R. M., Virgilio, R. W., and Brimm, J. E. Hypertonic sodium lactate versus lactated Ringer's solution for intravenous fluid therapy in operations on the abdominal aorta. *Surgery* **94:** 41, 1983.
5. Velasco, I. T., Pontieri, V., Rocha-e-Silva, M., and Lopes, O. U. Hyperosmotic NaCl and severe hemorrhagic shock. *Amer. J. Physiol.* **239:** H664, 1980.
6. Nakayama, S., Sibley, L., Gunther, R. A., Holcroft, J. W., and Kramer, G. C. Small volume resuscitation with hypertonic saline (2400 mOsm/liter) during hemorrhagic shock. *Circ. Shock* **13:** 149, 1984.
7. Nerlich, M., Gunther, R. A., and Demling, R. H. Resuscitation from hemorrhagic shock with hypertonic saline or lactated Ringer's (effect on the pulmonary and systemic microcirculations). *Circ. Shock* **10:** 179, 1983.
8. Baue, A. E., Tragus, E. T., and Parkins, W. M. A comparison of isotonic and hypertonic solutions and blood on blood flow and oxygen consumption in the initial treatment of hemorrhagic shock. *J. Trauma* **7:** 743, 1967.
9. Gazitua, S., Scott, J. B., Chou, C. C., and Haddy, F. J. Effect of osmolarity on canine renal vascular resistance. *Amer. J. Physiol.* **217:** 1216, 1969.
10. Lundvall, J., Mellander, S., and White, T. Hyperosmolarity and vasodilation in human skeletal muscle. *Acta Physiol. Scand.* **77:** 224, 1969.
11. Nakayama, S., Kramer, G. C., Carlsen, R. C., and Holcroft, J. W. Infusion of very hypertonic saline to bled rats: Membrane potentials and fluid shifts. *J. Surg. Res.* **38:** 180, 1985.
12. Thoren, L. Dextran as a plasma volume substitute. In G. A. Jamieson and T. S. Greenwalt (Eds.), *Blood Substitutes and Plasma Expanders.* New York: Liss, 1978.
13. Powers, S. R., Shah, D., Ryon, D., Newell, J., Ralph, C., Scovill, W., and Dutton, R. Hypertonic mannitol in the therapy of the acute respiratory distress syndrome. *Ann. Surg.* **185:** 619, 1977.
14. McNamara, J. J., Mills, D., and Aaby, G. V. Effect of hypertonic glucose on hemorrhagic shock in rabbits. *Ann. Thorac. Surg.* **9:** 116, 1970.
15. Rowe, G. G., McKenna, D. H., Jaramillo, C. V., and Crumpton, C. W. The systemic and coronary hemodynamic effect of sodium bicarbonate. *Amer. J. Med. Sci.* **248:** 424, 1964.
16. deFelippe, J., Jr., Timoner, J., Velasco, I. T., Lopes, O. U., Rocha e Silva, M., Jr. Treatment of refractory hypovolemic shock by 7.5% sodium chloride injections. *Lancet* **2:** 1002, 1980.
17. Gibson, J. G., and Evans, W. A., Jr. Clinical studies of the blood. Vol. I: Clinical application of a method employing the azo dye 'Evans Blue' and the spectrophotometer. *J. Clin. Invest.* **16:** 301, 1937.
18. Traverso, L. W. Personal communication.

jsm/

Small Volume Resuscitation with Hypertonic Saline Dextran Solution

George C. Kramer, Ph.D.

Paul R. Perron, B.S.

D. Craig Lindsey, B.S.

Hung Sy Ho, B.S.

Robert A. Gunther, Ph.D.

Walter A. Boyle, M.D.

James W. Holcroft, M.D.

Departments of Human Physiology, Surgery and Anesthesiology,
School of Medicine, University of California at Davis, CA 95616

Running Title: Small volume hypertonic resuscitation

Supported by Department of Defense contract No. DAMD 17-84-C-3175

Address reprint requests to: George C. Kramer
Dept. of Human Physiology
University of California
Davis, CA 95616
(916) 752-2180

ABSTRACT

Small volume hypertonic resuscitation has been proposed as an effective means for restoration of cardiovascular function after hemorrhage at the scene of an accident. We evaluated the cardiovascular, metabolic and neurohumoral response of resuscitation after hemorrhage using 200 ml of 2400 mosm sodium chloride-6% dextran 70. Unanesthetized adult sheep were bled to maintain mean arterial pressure at 50 mm Hg for 3 hrs, shed blood volume = 42 ± 7 ml/kg. Then sheep were treated with a single bolus infusion of hypertonic saline dextran (n = 7) or normal saline, (control group, n = 7) and then observed for a 30 minute period of simulated patient transport during which no additional fluid was given. Hypertonic saline dextran caused rapid restoration of blood pressure and cardiac output within 2 minutes of infusion. Cardiac output remained at or above baseline, while both O_2 consumption and urine output increased to above baseline during the 30 minutes of simulated patient transport. By comparison 200 ml of normal saline caused only a small increase in blood pressure and no improvement in cardiac output or oxygen consumption. After this 30 minute period both groups were given lactated Ringers as needed to return and maintain cardiac output at its baseline value. The volume of lactated Ringers required to maintain cardiac output was less in the hypertonic group, 371 ± 168 ml, only 1/6 that of the control group, 2200 ± 814 ml. In summary, after 3 hours of hypovolemia a small volume of hypertonic saline dextran, about 4 ml/kg, fully restored cardiovascular and metabolic function for at least 30 minutes and significantly lowered the total volume requirements of resuscitation.

INTRODUCTION

The volumes of resuscitation fluid required to restore cardiovascular function after hemorrhage and trauma are large, perhaps 2-3x shed blood volume. Optimal therapy would be prompt infusion of adequate volume. Unfortunately, in urban areas, infusion therapy administered at the scene of an accident and during transit by paramedics is often inadequate, with only 500-1000 ml of fluid being infused before hospital arrival (1). This has led many trauma surgeons, including ourselves, to propose a policy of "scoop and run" for trauma victims, i.e. immediate patient transport and volume resuscitation initiated upon arrival at the emergency room. On the other hand, field therapy would appear to be justified if effective restoration of cardiovascular function could be accomplished by paramedical personnel.

Infusions of very hypertonic solutions may provide a means for effective field resuscitation. Hypertonic sodium chloride solution, 2400 mosm, has been shown to cause rapid improvement in blood pressure even when given in very small volumes, 4 ml/kg, to dog (2,3), sheep (4,5), pig (6), rat (7) and man (8). This restoration of cardiovascular function has been attributed to a neurogenic reflex (3,9), but it also may result from an osmotically induced redistribution of intracellular water into the extracellular space (4,5).

Hyperosmolality per se, and not the type of solute, appears responsible for the near immediate improvement in cardiovascular function in hypovolemic animals. Smith et al (5) examined five different solutes each at a concentration of 2400 mosm in resuscitating hypovolemic sheep. All solutions were equally effective in initially restorating arterial pressure and cardiac output. This improvement was only transitory, however, unless a hyperoncotic colloid, 6% dextran 70, was added to the infusion solution. The hypertonic saline resulted in redistribution of body water from the intracellular to the extracellular

space; while the colloid resulted in further redistribution into the vascular compartment.

In the present study we further evaluated hypertonic saline dextran resuscitation by measuring cardiovascular, metabolic and hormonal responses of adult unanesthetized sheep during hemorrhagic hypotension and after infusions of 200 ml of hypertonic saline dextran. The volume chosen was small enough so that it could be easily administered in the field or during a short ambulance ride. After a simulated transport time of 30 minutes, the sheep were then given conventional large volume isotonic resuscitation. The purpose of these experiments was to mimic field and hospital conditions in resuscitating a patient in hemorrhagic shock. The study addressed the following questions: Can a small volume of hypertonic saline dextran fully restore and sustain normal cardiovascular and metabolic function after hemorrhage and during simulated patient transport time? What are the consequences of following small volume field resuscitation with traditional isotonic resuscitation? What are the electrolyte changes and total volume requirements in such a clinical situation?

METHODS

Animal Preparations

Seven adult female sheep, 43-55 kg, were anesthetized with halothane/nitrous oxide and surgically prepared with chronic cannulations of the thoracic aorta and superior vena cava using silastic catheters placed through a neck incision. A Swan-Ganz thermodilution catheter was placed to allow monitoring of central venous pressure, pulmonary artery pressure and cardiac output. Studies were performed 3-5 days after surgery. On the day of an experiment a Foley catheter was inserted into the bladder for urine collection.

Experimental Protocol

All experiments were conducted on unanesthetized animals kept unrestrained in cages. Baseline measurements of pressure and blood sampling were obtained over a two hour period. The sheep were then bled from their venous catheter until mean arterial pressure was lowered to 50 mm Hg over 5 - 15 minutes. Blood pressure was maintained between 45-55 mm Hg for 3 hrs by additional bleeding as required.

After 3 hours of hypotension each sheep was given a 1-2 minute i.v. infusion of 200 ml of hypertonic saline (1.2 N), 6% dextran 70 (Macrodex, Pharmacia), or normal saline by random selection. No further treatment was given during 30 minutes of simulated patient transport time. After this 30 minute period, sheep were given lactated Ringers as required to return and maintain cardiac output at its baseline value. Infusion rate was adjusted by a sliding scale from 0-200 ml/min, depending on the deficit in cardiac output.

After the 2 hr infusion of Ringers solution data collection was stopped and all shed blood was returned through a filtered blood administration set. The protocol was repeated seven to ten days after the first experiment, except the alternate solution was used. In all, seven experiments were performed with hypertonic resuscitation and seven with normal saline in a total of seven sheep.

Measurements

Aortic, central venous, and pulmonary artery pressures were measured with pressure transducers and continuously recorded on a strip chart recorder. Cardiac output was measured using an Edwards Cardiac Output Computer. Blood gases were measured with an Instrumentation Laboratories Blood Gas Analyzer. Hemoglobin was measured with a Coulter Hemoglobinometer. Urine was collected in a closed drainage bag; volume was determined every 30 min using a graduated cyclinder. Hematocrit was measured with microhematocrit tubes. Serum sodium

and potassium were measured on an Instrumentation Laboratories Flame Photometer. Osmolality was measured by freezing point depression with an Advanced Instruments osmometer. Oxygen consumption (ml/min) was calculated as the difference between arterial and mixed venous oxygen content multiplied by cardiac output. Glucose, lactate and blood urea nitrogen were measured by colorimetric enzymatic assays. Arginine vasopressin was measured by radioimmunoassay; catecholamines were measured by radioenzymatic assay. Total peripheral resistance (TPR) was calculated as the difference of mean arterial and central venous pressures divided by cardiac output.

Statistics

The paired Student's t test was used to compare variable differences between the two solutions used on each sheep. The Bonferroni Method was used to modify the t-test for multiple comparisons. Differences were considered significant when $P < 0.05$.

RESULTS

Data was grouped into Table 1 - Cardiovascular Variables, Table 2 - Blood and Plasma Solutes, Table 3 - Vasopressin and Catecholamines. All data shown are means ± 1 standard deviation of seven experiments each for both the hypertonic/hyperoncotic group and the control group given normal saline.

Baseline values for the two groups were similar. We have previously found that conducting 2 experiments 7 - 10 days apart on the same sheep is a sensitive and reproducible model to compare different resuscitation regimens (4,5,10). The shed blood volume required to maintain 3 hrs of hemorrhagic hypotension was 42.8 ± 8.0 ml/Kg for the first experiment, while it was 41.5 ± 7.5 ml/Kg for the second experiment. Three sheep were given hypertonic saline dextran first; four sheep were given normal saline first.

Hemorrhage:

Sheep were bled through their venous lines and required 5-15 minutes to reduce arterial pressure to 50 mm Hg. Typically 30-35 ml/kg of blood were removed during the first 30 minutes. After 3 hours mean shed blood volume was 42.0 ± 6.5 ml/kg for hypertonic saline dextran group and 42.0 ± 9.0 ml/kg for the control. This is about 2/3's of the sheep's estimated blood volume of 66 ml/kg (11). Cardiovascular, metabolic and neurohumoral variables were similar for the two groups during both baseline and hemorrhage. Cardiac output was reduced to 45-50% throughout hemorrhage, Fig. 2. Total peripheral resistance increased 40% and central venous pressure decreased 4-6 mm Hg, Table 1. Oxygen consumption decrease 12-30%, Fig. 3. and lactate increased 6-12x, Table 2. Despite this metabolic acidosis, pH remained normal due to respiratory compensation. Urine output decreased to 1/3 of baseline, Fig. 4. Arginine vasopressin increased 30-60 fold while norepinephrine and epinephrine were increased 6-20 fold, Table 4.

Small Volume Resuscitation and Simulated Patient Transport:

The 200 ml of hypertonic saline dextran or normal saline was given as a bolus infusion into the right atrium over 2 minutes. In the hypertonic group, blood pressure began to rise within 30 seconds from the beginning of infusion. By the end of the 2 minute infusion arterial pressure was restored to normal. Cardiac output and blood pressure at 3 minutes after bolus had increased to above baseline, Figures 1 and 2. After hypertonic resuscitation total peripheral resistance returned to baseline; central venous pressure increased, but was still less than baseline, Table 1. An equal volume of normal saline improved mean arterial pressure 10-20 mm Hg but had very little effect on cardiac output.

Metabolic improvement was noted 30 minutes after hypertonic resuscitation as oxygen consumption increased to above baseline, indicating early payback of

the oxygen debt incurred during hemorrhage, Fig. 3. Likewise mean serum lactate decreased with hypertonic saline dextran, but not with normal saline. A robust diuresis developed within 10 minutes of the bolus of hypertonic saline; after 30 minutes urine flow rate was 5x baseline levels. Improved cardiovascular function at baseline or better was maintained in all sheep through the entire 30 minutes of simulated patient transport time.

Lactated Ringers Infusion during Simulated Hospital Resuscitation:

In the control group, the cardiac output remained at hemorrhage levels or 50% of baseline during the 30 minutes of simulated patient transport. Thus Ringers resuscitation was usually begun at a rate of 200 ml/min in this group. Only 1 sheep in the hypertonic group had a cardiac output below baseline, and it was reduced only 10%. Figure 5 shows the cumulation volumes of fluid required in both groups to maintain cardiac output at baseline. Cardiac output and blood pressure were easily maintained with Ringers resuscitation in both groups. Substantially more fluid, 4-7x, was required in the control group 2200 ± 814 ml versus 371 ± 168 ml in the hypertonic group, Figure 4. The post hemorrhage elevations in vasopressin and catecholamines were similarly reduced during resuscitation except that vasopressin was higher after 2 hrs of Ringers resuscitation in the hypertonic group, Table 3. During Ringer's resuscitation the cardiovascular and metabolic variables were similar in both groups with no apparent differences.

DISCUSSION

The rapid improvement in blood pressure and cardiac output after hypertonic resuscitation may result from multiple mechanisms. Using hypertonic saline alone, Rocha e Silva's group and others have shown that acutely bled anesthetized dogs had rapid and sustained normalization of blood pressure and cardiac

output after 4 ml/kg of 2400 mosm hypertonic saline without any additional fluid replacement (2,3,9). Their studies indicated that a vagal mediated pulmonary reflex dilated peripheral resistance vessels while constricting venous capacitance vessels (3,12). They reported little expansion of blood or plasma volume after hypertonic resuscitation. On the other hand, Smith et al found that bled unanesthetized sheep had only a transient improvement in cardiac output after hypertonic saline alone and required the addition of a hyperoncotic colloid for a sustained response (5). In Smith's studies, about 4 ml/kg of hypertonic saline rapidly increased measured plasma volume, 10-12 ml/kg. This volume expansion was sustained only when dextran 70 was added to the same infused volume.

We cannot rule out a role for a vagal reflex in our studies, but volume expansion due to the hyperosmotic/hyperoncotic saline-dextran mixture must be a major contributing factor. We can estimate how much cellular water is drawn out by hypertonic resuscitation in a 50 kg sheep by assuming intracellular water equals 30-40% of body weight and by assuming that there is no net transport of osmoles across the cell membrane. Plasma osmolality increased from 313 mosm/l at end of hemorrhage to 334 mosm/l after hypertonic resuscitation, Table 2. This would cause a 6% loss of intracellular water or a total of 940-1260 ml of water transfered from the cellular to extracellular compartments. The control group required over 1800 ml more fluid during Ringers resuscitation suggesting additional mechanisms are operative. Much of the greater efficiency may be attributed to the hyperoncotic dextran which causes a greater fraction of extracellular fluid to be partitioned in the vascular space. The measured decrease in Hb suggests a 33% increase in plasma volume. Smith et al, found a 37% increase in measured plasma volume when a similar volume was given after a 2 hour hemorrhage.

The improved cardiovascular and renal function after hypertonic resuscitation did not appear to be mediated by increases in circulating epinephrine or dopamine, while the decreases in vasopressin and norepinephrine may partially account for the decreased peripheral resistance following hypertonic resuscitation. After hypertonic saline dextran the sharp fall in vasopressin suggests that increased blood volume inhibited vasopressin release more than increased osmolality stimulated release, whereas the significance between the groups at the end of the study is probably due to the osmolality difference.

The other apparent beneficial effects of hypertonic resuscitation may have been secondary to return of blood pressure and cardiac output or result from other specific mechanisms. Total peripheral resistance decreased to below baseline which may be attributed to a reflex (3) or a direct vasodilatory effect of hyperosmolality on vascular smooth muscle (13,14). Increased urine output may result from a combination of improved renal blood flow (14) as well as release of atrial naturetic factor (15). Overall metabolic improvement probably resulted from greater oxygen delivery secondary to increased cardiac output.

In summary, small volume resuscitation with about 4 ml/kg of 2400 mosm saline-6% dextran 70 rapidly normalized cardiovascular and metabolic function after 3 hrs of hemorrhage. This response was well sustained over a 30 minute period with no additional fluid, and over a 2-1/2 hr period with only an additional 6 ml/kg of isotonic fluid. The high efficiency of hypertonic resuscitation may provide paramedics, military corpsmen, and search and rescue teams with a physiological solution to the problem of field resuscitation of hemorrhage and trauma.

REFERENCES

1. Smith JP, Bodai BI, Hill AS: Prehospital stabilization of critically injured patients: a failed concept. J Trauma 25:65-70, 1985.

2. Velasco IT, Pontieri V, Rocha e Silva M, and Lopes OU: Hyperosmotic NaCl and severe hemorrhagic shock. Am J Physiol 239:H664-H673, 1980.

3. Lopes OU, Pontieri V, Rocha e Silva M Jr, Velasco IT: Hyperosmotic NaCl and severe hemorrhagic shock: role of the innervated lung. Am J Physiol 241:H883-H890, 1981.

4. Nakayama S, Sibley L, Gunther RA, Holcroft JW, Kramer GC: Small volume resuscitation with hypertonic saline (2400 mOsm/liter) during hemorrhagic shock. Cir Shock 13:149-159, 1984.

5. Smith GJ, Kramer GC, Perron P, et al: A comparison of several hypertonic solutions for resuscitation of bled sheep. J Surg Res 39:517-528, 1985.

6. Traverso LW, Bellamy RF, Hollengach SJ, et al: Hypertonic sodium chloride solutions: Effect on hemodynamics and survival after hemorrhage in swine. (in press) J Surg Res.

7. Nakayama S, Kramer GC, Carlsen RC, Holcroft JW: Infusion of very hypertonic saline to bled rats: Membrane potentials and fluid shifts. J Surg Res 38: 180-186, 1985.

8. deFelippe J Jr, Gimoner J, Velasco IT, Lopes OU, Roche e Silva M Jr: Treatment of refractory hypovolemic shock by 7.5% sodium chloride injections. Lancet 2:1002-1004, 1980.

9. Younes RN, Aun F, Tomida RM, et al: The role of lung innervation in the hemodynamic response to hypertonic sodium chloride solutions in hemorrhagic shock. Surgery 1985; 98:900-906.

10. Nerlich M, Gunther RA, Demling RH: Resuscitation from hemorrhagic shock with hypertonic saline or lactated Ringer's (effect on the pulmonary and systemic microcirculations). Circ Shock 10:179-188, 1983.

11. Hodgetts VE: The dynamic red cell storage function of the spleen in sheep. Aust J Exp Biol 39:187-196, 1961.

12. Lopes OU, Velasco IT, Guertzenstein PG, Rocha e Silva M Jr, Pontieri V: Hypertonic NaCl restores mean circulatory filling pressure in severely hypovolemic dogs (in press) Hypertension.

13. Lundvall J, Mellander S, White T: Hyperosmolarity and vasodilation in human skeletal muscle. Acta Physiol Scand 77:224-233, 1969.

14. Gazitua S, Scott JB, Chou CC, Haddy FJ: Effect of osmolarity on canine renal vascular resistance. Am J Physiol 217:1216-1223, 1969.

15. Richards, AM, Nicholis MG, Ikram H, Webster MWI, Yandle TG, Espiner EA: Renal hemodynamic and hormonal effects of human alpha atrial natriuretic peptides in healthy volunteers. Lancet:I:545-549, 1985.

TABLE 1: Cardiovascular Variables

	HR (min^{-1})		CVP (mm Hg)		TPR (mm Hg·L^{-1}·min)		SV (ml)		PAP (mm Hg)		RR (min^{-1})	
	NS	HS	NS	HS	NS	HS	NS	HS	NS	HS	NS	HS
Baseline	95 ±22	101 ±23	0 ± 2	0 ± 1	16 ± 3	17 ± 2	62 ±19	57 ±18	15 ± 3	14 ± 5	31 ±14	38 ±18
Hemorrhage (min)												
30	82 ±32	87 ±51	-5 ± 3	-4 ± 4	20 ± 6	21 ± 4	37 ±10	37 ±15	9 ± 3	9 ± 4	57 ±23	54 ±26
120	116 ±46	131 ±44	-5 ± 3	-5 ± 4	22 ± 5	21 ± 6	25 ± 8	23 ±10	9 ± 4	10 ± 4	50 ±25	71 ±34
180	128 ±41	139 ±65	-6 ± 4	-6 ± 2	25 ±10	22 ± 5	20 ± 7	21 ± 8	9 ± 3	9 ± 4	87 ±38	65 ±37
Post Bolus (min)												
3	131 ±45	169 ±37	-5 ± 6	-2 ± 8	24 ± 5	13* ± 4	25 ± 8	43* ± 7	9 ± 5	13 ± 7	38 ± 4	21* ± 4
10	152 ±39	161 ±50	-6 ± 3	-4 ± 6	23 ± 4	14* ± 4	22 ± 7	40* ± 7	9 ± 4	14 ± 6	37 ±22	20 ± 3
30	151 ±41	186 ±27	-7 ± 4	-4 ± 4	23 ± 7	16* ± 4	21 ± 9	32* ± 6	11 ± 3	14 ± 7	39 ±24	19 ± 2
Resuscitation (min)												
10	164 ±37	186 ±28	-6 ± 5	-4 ± 5	19 ± 4	16 ± 4	33 ±10	32 ± 7	15 ± 3	15 ± 5	22 ± 7	18 ± 3
60	146 ±35	167 ±28	-6 ± 4	-4 ± 4	16 ± 5	17 ± 4	43 ±18	33 ± 5	15 ± 5	14 ± 4	22 ± 7	19 ± 2
120	138 ±30	149 ±28	-5 ± 4	-2 ± 3	15 ± 2	15 ± 3	46 ±14	41 ± 7	15 ± 4	16 ± 2	21 ± 2	18 ± 2

Mean ± standard deviation shown for:

Heart rate (HR), central venous pressure (CVP), total peripheral resistance (TPR), stroke volume (SV), pulmonary artery pressure (PAP) and respiratory rate (RR).

* $P < 0.05$ HSD vs NS.

TABLE 2: Blood and Plasma Solutes

	Hct		Hb g/100 ml		Osmolality mosm/l		$[Na^+]$ meq/l		$[K^+]$ meq/l		$[Cl^-]$ meq/l		[Glucose] mmol/l		[BUN] mmol/l		[Lactate] mmol/l	
	NS	HSD	NS	HSD	NS	HSD	NS	HSD	NS	HSD	NS	HSD	NS	HSD	NS	HSD	NS	HSD
Baseline	24.3	23.7	8.7	8.5	303	303	152	150	3.9	3.8	112	111	4.9	5.4	7.1	8.5	0.7	0.6
	±3.2	±2.5	±1.2	± 1	± 5	± 5	± 2	± 3	±0.4	±0.3	± 2	± 3	±1.7	±1.9	±1.9	±1.4	±.2	±.3
Hemorrhage (min)																		
60	21.6	21.0	7.3	6.5	310	308	153	152	4.5	4.3	110	110	6.8	6.9	8.4	9.4	2.3	2.9
	±1.6	±2.3	±1.0	±2.8	± 9	± 5	± 3	± 3	±0.5	±0.4	± 2	± 3	±3.6	±4.0	±2.4	±1.4	±1.3	±1.2
120	21.5	21.0	7.4	7.2	311	310	154	153	4.2	4.4	110	106	7.8	8.6	8.6	10.2	3.0	4.8
	±1.9	±1.5	±1.1	±0.6	± 6	± 4	± 3	± 4	±0.7	±0.6	± 2	± 6	±4.1	±3.9	±2.6	±2.0	±1.7	±2.1
180	21.0	20.0	7.7	6.8	313	313	154	153	4.2	4.6	109	107	10.1	10.5	8.9	10.8	4.7	7.9
	±4.5	±2.3	±1.8	±0.8	± 8	± 6	± 4	± 3	±0.6	±0.9	± 1	± 3	±5.6	±5.6	±2.6	±2.1	±2.8	±5.4
Post Bolus (min)																		
15	20.0	13.0*	6.5	5.1*	311	334*	154	165*	3.5	3.4	112	124*	10.6	9.2	8.8	10.7	4.5	7.3
	±3.9	±1.8	±1.3	±1.5	± 7	± 4	± 3	± 3	±0.6	±0.5	± 2	± 3	±4.9	±4.4	±2.5	±2.2	±3.1	±5.6
30	20.0	14.0*	6.6	4.9*	311	333*	154	164*	3.6	3.3	113	122*	10.1	9.0	8.9	10.3	4.5	6.2
	±4.0	±1.6	±1.6	±2.5	±10	± 6	± 4	± 3	±0.4	±0.6	± 2	± 4	±4.9	±4.4	±2.5	±1.9	±2.9	±5.4
Resuscitation (min)																		
10	15.5	15.5	5.3	5.1	312	325	153	162*	3.4	3.3	113	122*	9.0	8.7	8.3	10.2	4.8	5.7
	±3.0	±3.0	±1.0	±0.6	±10	± 6	± 4	± 4	±0.6	±0.5	± 2	± 4	±4.4	±4.0	±2.3	±1.8	±3.2	±5.1
60	14.5	14.0	5.3	5.3	304	323	153	162*	3.0	3.3	109	117*	7.9	7.7	8.3	9.8	3.3	3.5
	±2.7	±2.0	±4.2	±1.0	± 8	± 6	± 4	± 3	±0.3	±0.7	± 7	± 8	±4.2	±3.3	±2.0	±1.7	±2.6	±3.2
120	14.0	13.0	4.8	4.8	305	320	153	161*	3.1	3.3	115	120*	6.9	6.3	7.7	9.4	2.4	2.4
	±2.9	±1.6	±1.0	±0.8	± 7	± 6	± 4	± 2	±0.3	±0.6	± 2	± 4	±3.6	±2.4	±2.0	±1.6	±1.6	±1.9

Mean ± standard deviation shown for:

* $P < 0.05$ HSD vs NS

Hematocrit (Hct), Hemoglobin (Hb), osmolality and plasma concentrations of sodium, potassium, chloride, glucose, blood urea nitrogen (BUN) and lactate

TABLE 3

Vasopressin and Catecholamines

	Arginine Vasopressin pg/ml		Epinephrine pg/ml		Norepinephrine pg/ml		Dopamine pg/ml	
	NS	HSD	NS	HSD	NS	HSD	NS	HSD
Baseline	7 ± 3	5 ± 2	63 ± 21	62 ± 24	240 ±133	332 ±193	70 ± 56	60 ± 58
Hemorrhage (min)								
30	451 ±537	232 ±141	352 ±250	336 ±359	338 ±196	502 ±138	126 ±123	139 ±132
180	365 ±485	273 ±188	536 ±262	1301 ±1714	2045 ±2060	1805 ±1296	137 ±102	229 ±259
Post Bolus (min)								
15	134 ±183	74 ±22	234 ±148	150 ±144	803 ±474	666 ±539	50 ±25	102 ±127
Resuscitation (min)								
30	51 ±61	46 ± 6	152 ±107	152 ±128	648 ±458	715 ±410	241 ±505	89 ±94
120	13 ± 6	32* ± 7	75 ±25	102 ±72	421 ±187	460 ±246	116 ±152	123 ±181

* $P < 0.05$

FIGURE LEGENDS

FIGURE 1: Arterial pressure was quickly restored after 3 hrs of hemorrhagic hypotension by infusion of 200 ml of hypertonic saline dextran.

FIGURE 2: Cardiac output is increased to above baseline and remains elevated through 30 minutes of simulated patient transport.

FIGURE 3: Oxygen consumption improved to above baseline in all seven sheep treated with 200 ml of hypertonic saline dextran; 200 ml of normal saline had no effect on oxygen consumption.

FIGURE 4: Within 10 minutes of infusion of hypertonic saline dextran a robust diuresis began.

FIGURE 5: Volume of lactated Ringers required to restore and maintain cardiac output was 4-7x greater in control animals than in animals resuscitated with hypertonic saline dextran. Both groups started off with 200 ml of hypertonic or normal saline.

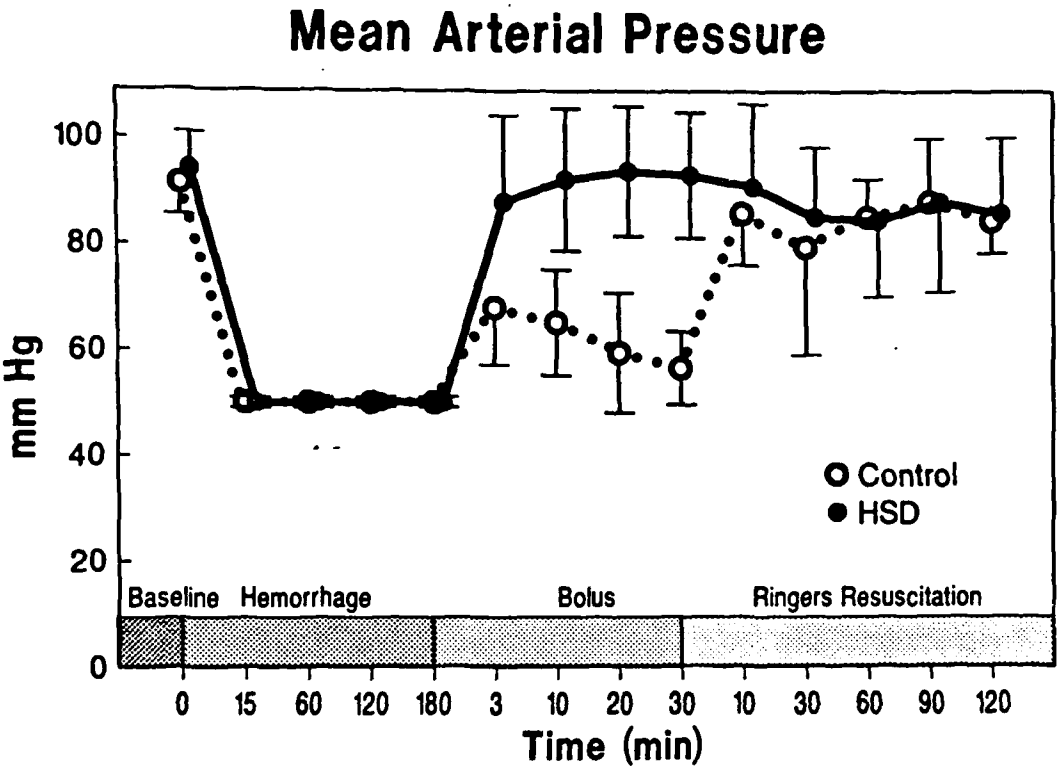

Mean Arterial Pressure
mm Hg
100
80
60
40
20
0
Control
HSD
Baseline
Hemorrhage
Bolus
Ringers Resuscitation
0
15
60
120
180
3
10
20
30
10
30
60
90
120
Time (min)

Fig 1

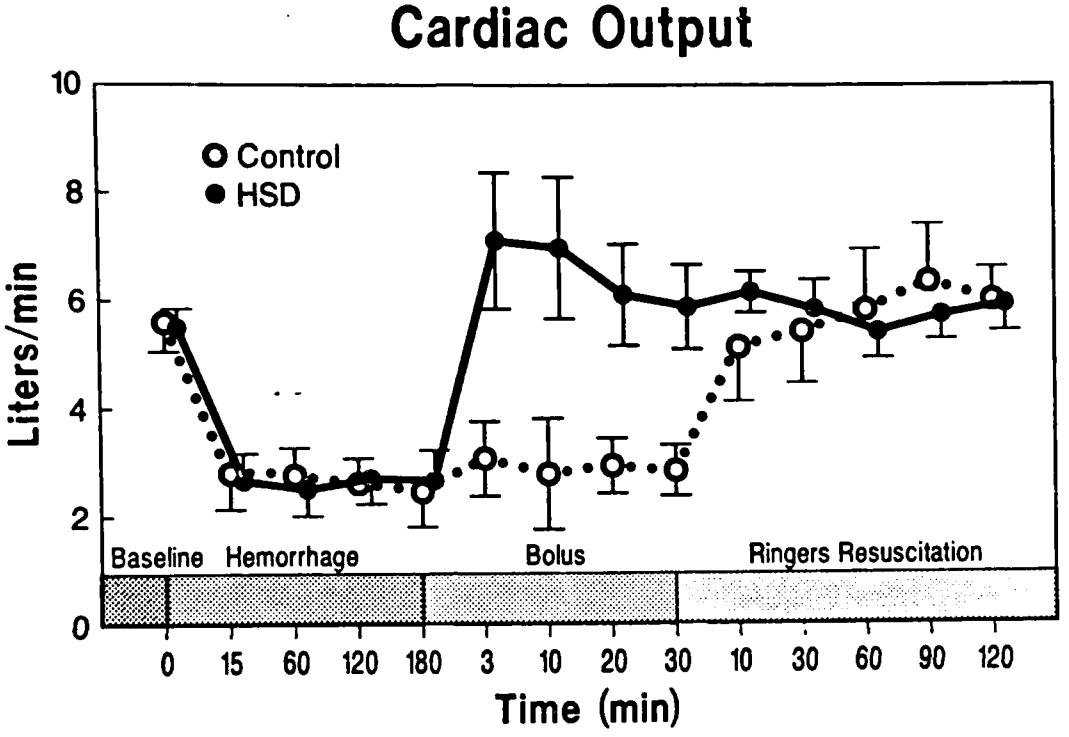

Cardiac Output
Control
HSD
Liters/min
10
8
6
4
2
0
Baseline
Hemorrhage
Bolus
Ringers Resuscitation
0
15
60
120
180
3
10
20
30
10
30
60
90
120
Time (min)

Fig 2

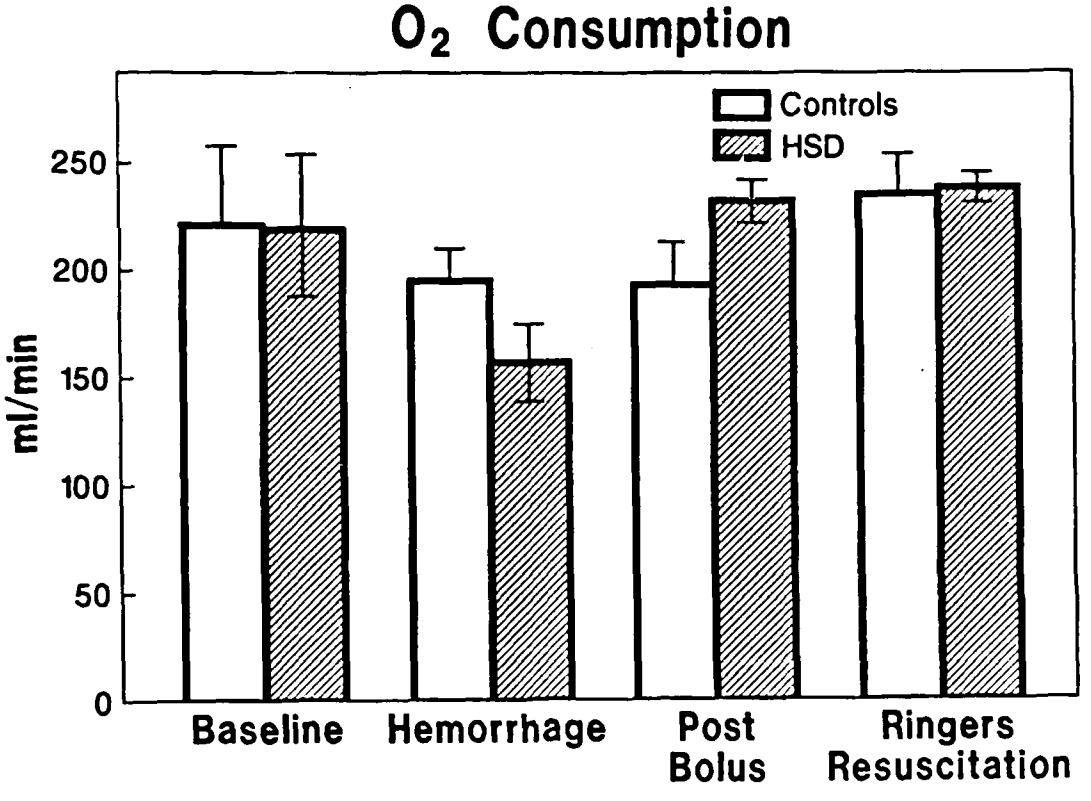

O_2 Consumption
Controls
HSD
ml/min
250
200
150
100
50
0
Baseline
Hemorrhage
Post Bolus
Ringers Resuscitation

Fig 3

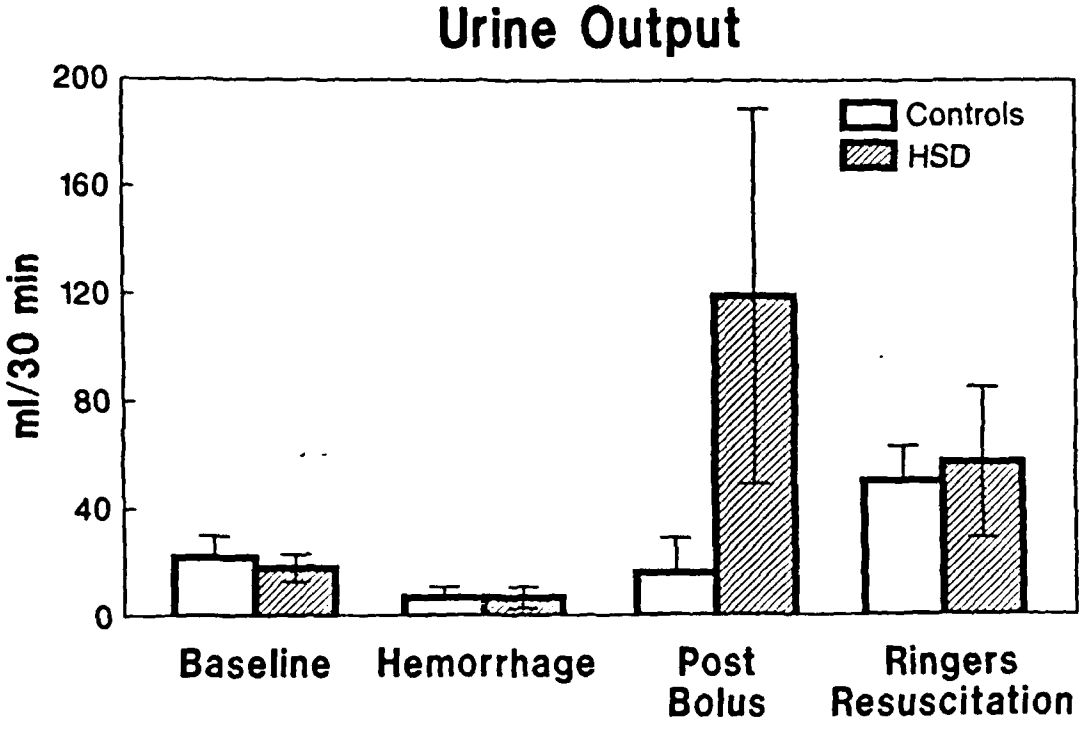

Urine Output
200
160
120
80
40
0
ml/30 min
Controls
HSD
Baseline
Hemorrhage
Post Bolus
Ringers Resuscitation

Fig 4

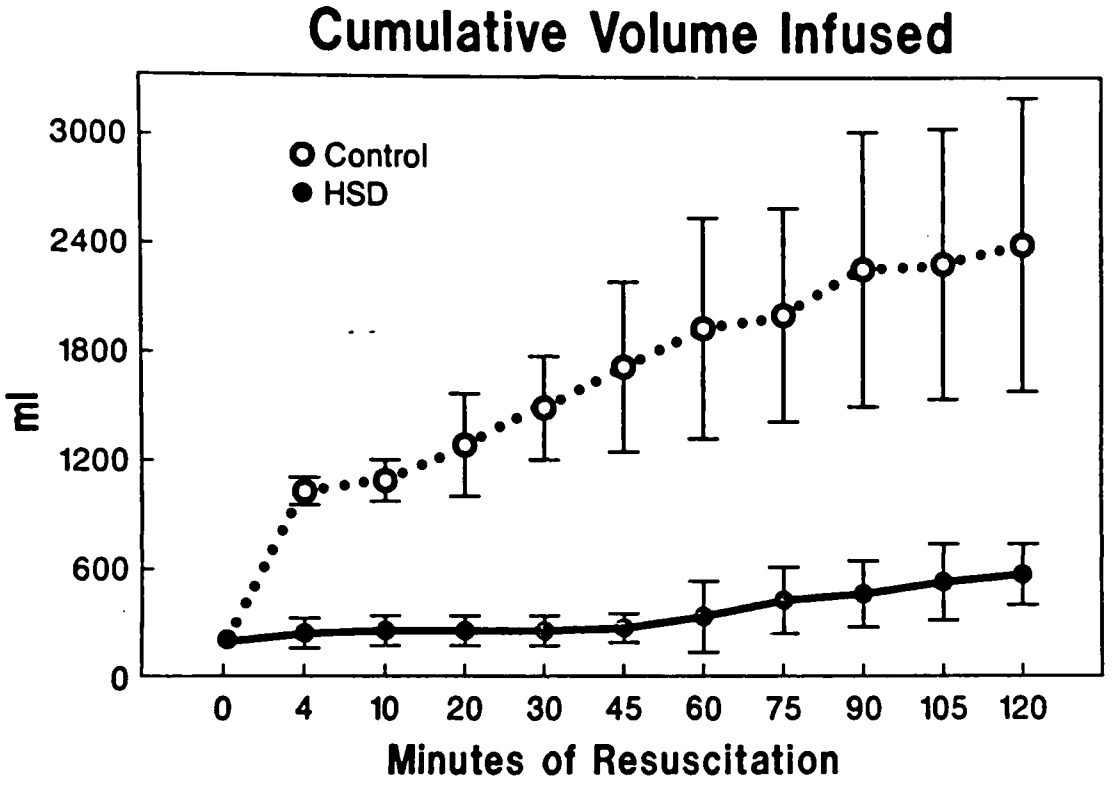

Cumulative Volume Infused
Control
HSD
ml
3000
2400
1800
1200
600
0
0
4
10
20
30
45
60
75
90
105
120
Minutes of Resuscitation

Fig 5

DISTRIBUTION LIST

4 copies	Commander Letterman Army Institute of Research (LAIR), Bldg. 1110 ATTN: SGRD-ULZ-RC Presidio of San Francisco, CA 94129-6815
1 copy	Commander U.S. Army Medical Research and Development Command ATTN: SGRD-RM1-S Fort Detrick, Frederick, Maryland 21701-5012
2 copies	Defense Technical Information Center (DTIC) ATTN: DTIC-DDAC Cameron Station Alexandria, VA 22304-6145
1 copy	Dean School of Medicine Uniformed Services University of the Health Sciences 4301 Jones Bridge Road Bethesda, MD 20814-4799
1 copy	Commandant Academy of Health Sciences, US Army ATTN: AHS-CDM Fort Sam Houston, TX 78234-6100


FILM